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Development of an accurate means to predict the nutritive value of wheat for broilers and an investigation of novel wheat factors on broiler performance

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1. Abstract

The aims of this project were to validate existing NIRS equations using commercially produced birds to provide a tool to predict the nutritive value of wheat for broilers and to investigate novel wheat factors (moisture content at harvest and microdochium content) which may influence broiler performance. Twenty six wheat samples were sourced from across Northern Ireland, Great Britain, Europe and North America and were formulated into broiler diets. The wheat samples were analysed for specific weight and *in vitro* viscosity and for a number of chemical constituents (gross energy, crude protein, total starch, microdochium, DON, ZON, ochratoxin A and B and amino acid contents). The majority of the relationships between the wheat chemical and physical parameters were weak and non-significant. However, there was a small, but significant relationship ($P<0.01$) between specific weight and microdochium content ($R^2=0.29$) Separate broiler trials were conducted at AFBI, Harper Adams University and Moy Park Ltd. to determine intake, liveweight gain (LWG) and feed conversion ratio (FCR). Results show differences in performance due to wheat sample. There was no significant relationship between wheat specific weight and broiler performance ($R^2=0.01$ for feed intake, $R^2=0.03$ for LWG and $R^2=0.02$ for FCR, respectively). However, wheat moisture content at harvest was shown to be positively related to LWG ($R^2=0.40$, $P=0.005$) which may be attributed to the positive effects of the drying process or the benefits of early-stage sprouting. A weak positive relationship was observed between wheat microdochium content and feed efficiency ($R^2=0.19$, $P<0.05$) but the reason for this is not understood. The effect of the mycotoxins DON and ZON were variable and difficult to explain.

NIRS equations were developed to predict wheat microdochium content and boiler performance. Wheat samples were scanned on a Foss NIRSystems 6500 spectrophotometer and analysed using Foss Chemometrics software Win ISI4. The strength of the relationship between actual and predicted microdochium content was reasonably robust (1-VR=0.70 for the entire dataset). However, the errors associated with the prediction and the ratio of prediction to deviation (RPD) were unacceptable. Although the errors and RPD values improved when the dataset was split into low, medium and high microdochium contents, they still did not reach an acceptable threshold. Nonetheless, this initial works shows that NIRS may have potential in predicting the microdochium content but the dataset needs to be increased to improve accuracy.

Strong NIRS prediction equations were developed to predict broiler performance ($R^2=0.92$, 0.83 and 0.85 for feed intake, LWG and feed efficiency, respectively) in trials at AFBI. The error associated with predictions were low (SEP=2.955, 2.787 and 2.023) and the RPD values indicated that the predictions were excellent, quantitative and good for feed intake, LWG and

feed efficiency, respectively. Using the equations to predict the performance of broilers within the Moy Park trial resulted in somewhat weaker relationships between actual and predicted performance (R^2=0.37, 0.46 and 0.46 for feed intake, LWG and feed efficiency, respectively). It is, therefore, concluded that while strong prediction equations have been developed using birds housed under commercial conditions, these equations could be further strengthened using additional samples.

2. Introduction

Studies in the scientific literature (Miller *et al* (2001) and Owens *et al* (2007)) report that there is a lack of relationship between the specific weight value of wheat and the feed nutritive value for grain. In the report by Owens *et al* (2007), the relationship is extremely weak and non-significant, with an R^2 value of 0.01 between specific wheat and apparent metabolisable energy. Despite this evidence, which is in agreement with the findings of many other research publications (e.g. Wiseman 2000), the industry continues to use specific weight as a measure of grain quality mainly because there is no accurate alternative and specific weight is a rapid and inexpensive test. However, due to the lack of relationship between specific weight and animal nutritive value, its continued use results in financial penalties for wheat producers. This is of particular importance in years of wet growing and harvesting conditions which have led to poor grain filling and subsequently low specific weight wheat and therefore perceived poor quality for the feed industry. Generalisations on the chemical composition of grain are made depending on the value of specific weight and several research reports have established that there are significant correlations between specific weight and chemical constituents. For example, Hickling (1994) found that there was a 0.5% decrease in starch content for each density point fall in specific weight of Canadian wheat with $R^2=0.93$. However, it must be highlighted that the majority of publications that report strong relationships between specific weight and chemical composition are based on small datasets. When a large dataset of 164 wheat samples were considered (Owens *et al* 2007), the relationships were found to be non-significant and as such, it can be concluded that there is no scientific justification for using specific wheat as an indication of either protein or starch content or on how a bird will perform. Therefore, the broiler industry urgently requires an accurate alternative to specific wheat to predict how wheat will perform and this project aimed to develop such an alternative.

There has been considerable research conducted on how chemical composition and other physical assessments of wheat relate to bird performance and energy utilisation but with contrasting results and little success. For example, Pirgozliev *et al* (2003) found that thousand grain weight was negatively correlated with feed conversion efficiency, whereas Hughes *et al* (1996) and Garnsworthy *et al* (2000) reported that it was not correlated with apparent metabolism energy. Owens *et al* 2007 found that there was a positive relationship between a combination of chemical and physical parameters and bird performance. The best correlation coefficient was 0.863 for feed efficiency but this involved the determination of nitrogen, thousand grain weight, rate of starch digestion, soluble non-starch polysaccharide content, neutral detergent fibre and dry matter, costing over £400 per sample and over 5 days of labour. Near infrared reflectance spectroscopy (NIRS) has been shown to be effective in predicting the nutritive value of forage for ruminants (Park *et al* 1998) and previous work by AFBI has indicated that it has the potential to accurately predict how a bird will perform when offered a diet based on particular wheat (Owens *et al* 2007). NIRS is a rapid, non-invasive procedure which, if proved to be commercially applicable, would be a viable alternative to specific weight.

Mycotoxins are toxic chemicals that are produced by fungi which infect crops. In wheat, mycotoxins can result from fungi contamination from field-borne infections (e.g. *Fusarium*) or from fungi contamination during storage (e.g. *Penicillium*). The most common F. mycotoxins are deoxynivalenol (DON) and zearalenonoe (ZON) and the most common P. mycotoxin is ochratoxin A. Alfatoxin B1, produced by *Aspergillus* fungi is another important mycotoxin and is currently the only one for which there are maximum permitted levels in animal feed due to the genotoxic effect in both animals and humans. Overall, the potential negative effect of mycotoxin contamination of broiler feed has been well documented and they can cause gastrointestinal disorders, reproductive failure, kidney damage, immune-suppression and reduction in production performance (http://www.food.gov.uk/policy-advice/mycotoxins/). However, in practice, their actual effect on broiler performance has not been well established (D'Mello and Macdonald, 1997) and there is a need to relate mycotoxin level with broiler performance using a large dataset of wheat. Andretta *et al* (2011) conducted a meta-analysis of 98 studies investigating the effect of specific mycotoxins on broiler performance and concluded that the magnitude of the response was still variable. There have been no studies conducted using large dataset of wheat and how the mycotoxin levels listed above influence broiler performance and nutrient digestibility. NIRS has been reported to be of potential use in predicting the mycotoxin level in wheat (e.g. Wrigley, 1999 and Girolamo, 2009). However, more work is needed to fully develop the prediction equations. Girolamo (2009) used Fourier Transform NIRS to develop a prediction equation for DON concentration in wheat and reported that a "limited to good" relationship was obtained using milled wheat. There is no other work reported in the open literature which has investigated the use of NIR to predict DON, ZON, alfatoxin B1 or ochratoxin A concentration in whole wheat grain.

Another fungi infection of wheat (which, in common with *Fusarium* species, also causes ear blight) is *Microdochium,* and in particular, *M. nivale*. While *microdochium* fungi do not produce any mycotoxins, there is a concern that their effect on the grain may reduce the nutritive value. Hare *et al* (1999) and Eken *et al* (2011) reported that grain infected with *microdochium* results in lower yields and also lowers the weight of individual grains. The relationship between microdochium concentration and broiler performance has never been investigated before and is a novel aspect of this study.

The use of exogenous enzymes in commercial poultry production is commonplace. Enzyme technology can provide a measurable and relatively consistent economic return on investment (Bedford 2000). Use of enzymes can reduce variation in animal performance improving feeding quality of feeding wheat. For example, poor quality wheat samples tend to show more improvement in feeding quality when supplemented with exogenous non-starch polysaccharide (NSP) degrading enzymes compared to good quality wheats (Bedford 2000). Other enzymes such as proteases, phytases and amylases may also offer value by improving the nutritional value of wheat for poultry. A further aim of this project was to examine the effect of exogenous enzyme supplementation to wheat based diets on broiler performance.

Project Objectives

1. To validate existing NIRS equations to provide a tool to predict the nutritive value of wheat for commercially produced broilers
2. To investigate the effect of moisture content at harvest and drying conditions of wheat on broiler performance and the use of enzymes on good and poor quality wheat.
3. To investigate the effect of mycotoxin and microdochium level on broiler performance.

3. Materials and methods

3.1. Study One – AFBI trial

Twenty wheat samples were sourced from across Northern Ireland (n=7), Republic of Ireland (n=2), Great Britain (n=8), USA (n=1), Germany (n=1) and France (n=1). The samples were analysed for specific weight, gross energy, crude protein, *in vitro* viscosity, total starch, microdochium, DON, ZON, ochratoxin A and ochratoxin B and amino acid contents. Specific weight was measured using a Digi-sampler hectolitre test weight machine (ELE International Limited), gross energy was determined using an automated bomb calorimeter and crude protein was determined using the Kjeldhal method. *In vitro* viscosity was determined using a modification of the method described by Bedford and Classen (1993). The digesta was centrifuged (6000rpm for 8min) and viscosity measured using a Brookfield LVDV II cone and plate viscometer at 20^{o}C. Total starch was determined using a Megazyme enzyme kit (Megazyme International Ireland Ltd). Microdochium content was determined by the Taqman probe method developed by Cockerell *et al* (2004). Mycotoxin analysis was conducted at QUB. Samples were extracted using QUECHERS (Quick Easy Cheap Effective Rugged Safe) method and purified with C18 silica sorbent before LC-MS/MS analysis.

Starter, grower and finisher broiler diets were formulated using each wheat sample. The diets were balanced for metabolisable energy, crude protein, lysine, methionine, threonine and tryptophan (Table 1). For each wheat sample, the inclusion of ingredients were adjusted slightly on the basis of the determined amino acid analysis of the wheat. (Appendix 1 shows the detailed diet formulations for all 20 wheat samples).

Table 1. Composition of experimental diets (AFBI trial)

	Starter	Grower	Finisher
Wheat (%)	60.3	65.2	71.5
Soyabean meal (%)	24.1	22.9	17.1
Full fat soya (%)	10.0	5.0	5.0
Soya oil (%)	1.92	3.73	3.81
Salt (%)	0.20	0.15	0.20
Sodium Bicarbonate (%)	0.32	0.28	0.25
DL Methionine (%)	0.18	0.14	0.08
Lysine HCl (%)	0.37	0.29	0.11
Threonine (%)	0.14	0.10	0.04
L-Tryptophan (%)	0.00	0.00	0.00
Limestone (%)	0.98	0.56	0.55
Mono dicalcium phosphate (%)	1.07	1.06	0.75
Minerals and vitamins (%)	0.50	0.50	0.50
Phytase (%)	0.02	0.02	0.02
Formulated analysis			
Crude protein (%)	23	21	19
Metabolisable energy (MJ/kg)	12.6	12.9	13.4
Lysine (%)	1.40	1.19	0.95

Two hundred male chicks (Ross 308) were obtained on day of hatch, weighed and divided into 20 weight blocks and arranged throughout the room according to a predetermined randomisation (Table 2) to give one pen per treatment. The starter diet was offered from day 1 to 14, the grower offered from day 14 to 21 and the finisher from day 21 to 35. Dry matter intake (DMI), liveweight gain (LWG), and feed conversion ratio (FCR) were determined for the starter, grower and finisher periods. This was repeated twice with a further 400 broilers to give a total of six pen replicates per treatment. Birds were offered feed and water *ad libitum*. Initial room temperature was 33°C. This was reduced by 1°C every day for the first week, and by 1°C every other day thereafter, to a minimum of 24°C. Light was provided for 18 h with the dark cycle being between midnight and 0600 h. The birds were fed *ad libitum* from 0-35 d. Food and drinking water were supplied from circular troughs and drinkers placed at floor level in the pens. Birds were weighed individually and the average pen weight was determined for each stage. At 21d, four birds (two heaviest and two lightest) were removed to maintain allowed stocking densities. Average pen weight was maintained.

Table 2. Randomisation of dietary treatment to pen number throughout broiler performance room

Pen	Run 1	Run 2	Run 3	Run 4	Run 5	Run 6
1	3	8	15	4	8	11
2	11	2	4	16	9	10
3	16	9	9	1	11	8
4	6	6	1	13	13	17
5	9	14	12	12	14	16
6	19	4	14	14	12	13
7	10	18	7	5	1	4
8	4	3	5	20	2	3
9	15	11	19	17	6	2
10	13	15	2	3	7	14
11	18	7	18	19	17	12
12	14	16	3	18	19	5
13	1	12	13	9	18	6
14	2	5	8	11	10	20
15	5	10	11	8	15	9
16	17	20	6	10	16	18
17	12	19	16	15	20	7
18	7	1	17	7	5	1
19	20	13	20	2	3	19
20	8	17	10	6	4	15

Simple regression analysis was performed to test the relationship between the chemical and physical parameters of the wheats. The results of the broiler trial were subjected to analysis of variance using Genstat. The trial was a randomised block design with six blocks and 20 diets. The data was analysed with initial weight as a covariate. Specific weights were known for the wheat used in all 20 diets and % moisture content at harvest was known for the wheat used in nine of the 20 diets. The data were firstly analysed regarding 20 diets as separate diets. They were then analysed using polynomial contrasts in order to determine whether differences between the diets were due to differences in the specific weight. They were then again analysed using a combination of factors and polynomial contrasts in order to determine whether differences between the diets were due to differences in the % moisture. The pen was taken as the experimental unit. Simple regression relationships between bird performance and chemical and physical parameters of wheat were established.

3.2. Study Two – Moy Park trial

Six wheat samples for commercial feed use were sourced during May to December 2014 from Denmark (old and new crop), Romania (old crop), Ireland (new crop) and the UK (old and new crop). A sufficient quantity of each wheat was placed in 1 tonne tote bags, labelled and stored in a secure warehouse until required for feed manufacture. Specific weight was determined and the wheat samples scanned on an Antaris II FT-NIR instrument to determine crude protein, ash, oil A, fibre and ash content.

Starter, grower, finisher and withdrawal diets were formulated using each wheat sample. The diets were balanced for metabolisable energy, crude protein and limiting amino acids (Table 3). For each wheat sample, the inclusion of ingredients was adjusted slightly on the basis of the determined CP content and calculated amino acid content using Evonik AminoDat4.0 regression equations (Appendix 2 shows the detailed diet formulations for all six wheat samples).

A total of 18,000 Ross 308 (as hatched) broiler chicks were obtained on day of hatch from a single Ross 308 breeding flock. Birds were allocated to 36 pens (500/pen) according to a pre-determined randomisation (Table 4) and start weight was balanced across pens. Feed intake, liveweight gain and FCR were determined weekly and over the entire period (38d). Bodyweight was measured at 7, 14, 21 and 28 days of age by weighing 50 birds per pen. Feed intake on a weekly basis was calculated by adding feed dispensed and the feed remaining in the feeders at the same intervals as the bodyweight measurement. Weekly FCR was then calculated using the feed and body weight measurements.

The results were analysed by Analysis of Variance using Genstat. The effects of wheat sample, location of production and old and new crop were tested for. Pen was taken as the experimental unit.

Table 3. Composition of experimental diets (Moy Park trial)

	Starter	Grower	Finisher	Withdrawal
Wheat (%)	59.9	60.0	66.3	66.0

GM Hipro Soya (%)	28.0	21.9	18.9	16.2
DDGS (%)	2.5	7.5	6.5	6.5
Whole Rapeseed (%)	3.0	4.5	0	5.5
Oil Blend (%)	2.4	2.5	4.8	3.0
Limestone (%)	0.85	1	1.05	0.9
Mono-Dicalcium Phosphate (%)	0.85	0.6	0.5	0.4
*Vitamin & Mineral Premix (%)	0.7	0.6	0.6	0.5
L-Lysine-HCl (%)	0.42	0.44	0.53	0.47
DL Methionine (%)	0.395	0.305	0.63	0.285
Commercial Additive (%)	0.3	0	0	0
Soyabean oil (%)	0.3	0.3	0	0
Sodium Carbonate (%)	0.25	0.25	0.2	0.15
Salt (%)	0.06	0.05	0	0.028
Threonine (%)	0.045	0.05	0.03	0.085
NSP Enzyme (%)	0.023	0.023	0.023	0.023
Phytase Enzyme (%)	0.01	0.01	0.01	0.01
Formulated analysis:				
Protein	22	20	18	18
Metabolisable energy Kcal/lb	12.78	13.14	13.42	13.47
Lysine	1.43	1.3	1.23	1.17

* The vitamin and mineral premix also contained threonine, sodium bicarbonate and anticoccidials.

Table 4. Randomisation of dietary treatment to pen number throughout broiler performance house (Moy Park trial)

Diet	Pen	Pen	Diet
5	19	18	3
3	20	17	1
4	21	16	5
6	22	15	2
1	23	14	4
2	24	13	6
5	25	12	1
6	26	11	2
4	27	10	3
2	28	9	5
1	29	8	6
3	30	7	4
4	31	6	2
6	32	5	3
1	33	4	4
3	34	3	5
5	35	2	6
2	36	1	1

3.3. Study Three – Harper Adams University trial

See Appendix 3 for the full study report.

3.4. Near Infrared Reflectance Spectroscopy (NIRS)

3.4.1. Development of performance prediction equations

The wheat samples used in all the trials were scanned as whole undried kernels on a Foss NIRSystems 6500 spectrophotometer. They were scanned in duplicate in a coarse transport quarter cell over the wavelength range 400-2498nm with readings taken at 2nm gaps. These scans of the new wheat samples along with the wheat scans from Owens *et al* 2007 were analysed using the Foss Chemometrics software Win ISI4. The mathematical treatment of standard normal variate and detrend (SNVD), first derivative, gap of 4 and smooth of 4 was applied. Modified partial least squares regression was performed on the data set on the range 400 nm – 2500nm and NIRS calibration and validation statistics generated to predict performance and compare with actual values obtained through the broiler trials.

3.4.2. Prediction of wheat microdochium content

The same NIRS scans and mathematical treatments applied as for Section 3.4.1. Calibration and validation statistics were generated to predict microdochium content and to compare with actual analysed microdochium content. The range of microdochium content was too wide (0.002-60.6pg/g) when the entire dataset was considered and so the dataset was split into low, medium and high categories and considered independently.

4. Results

4.1. Chemical and physical analysis of wheat used in the AFBI trial

The results of the chemical and physical analysis of the wheat samples are presented in Tables 5-8. Moisture content at harvest was recorded for nine of the 20 wheats used in the AFBI trial and ranged from 13.5% (wheat 16, Avator, GB) to 23% (wheat 1, Duxford, NI). The lowest IVV was observed in wheat 5, USA (6.86mPa.S) and the highest IVV observed in wheat 2, Grafton, NI (13.55mPa.S). There was a wide range in SW values with wheat 12, Neil Furniss, GB, having the lowest value of 55.7 kg/hL and wheat 5, USA having the highest value of 79.9 kg/hL. Gross energy of the wheats were similar with values ranging from 17.5 to 18.0 g/kgDM. The CP content of the wheat ranged from 90.2 g/kg (wheat 16, Avator, GB) to 128.4 g/kgDM (wheat 8, SA358, Germany). Wheat 17, Composite 1, NI, contained the lowest level of starch (567.2 g/kgDM) and wheat 11, Brockton, GB, contained the highest level of starch (640.7g/kgDM). Wheat microdochium content varied considerably with the lowest level detected in wheat 5, USA (0.45pg/ng) and the highest level detected in wheat 18, Composite 2, NI, (60.66pg/ng). DON was detected in all the wheat samples and ranged from 22.02 (wheat 6, 2, RoI) to 623.4 (wheat 13, Crusoe, GB). ZON was detected in nine of the 20 samples. Wheat 20, Composite 5, NI, contained the lowest level of ZON (3.26ng/g) and wheat 1, Duxford, NI, contained the highest level (38.4ng/g). Ochratoxin A was detected in wheat samples, 3 and 8 (10.57, 0.43 and 0.53ng/g respectively). Ochratoxin B was only detected in wheat 1, Duxford, NI (1.71ng/g) (Table 6).

The amino acid profile of the 20 wheats used for the AFBI trial are presented in Table 7. These profiles are summarised in Table 8, showing the minimum, mean, maximum and standard deviation values for each amino acid.

The majority of the relationships between the wheat chemical and physical parameters were weak and non-significant (Table 9). However, there was a weak (R^2=0.29) but significant (P=0.009) relationship between SW and microdochium content. Also, CP was found to be positively (R^2=0.55) and negatively (R^2=0.53) correlated with GE and starch content, respectively (P<0.001).

Table 5. Moisture content (MC) at harvest, *in vitro* viscosity (IVV), specific weight (SW), gross energy (GE) and crude protein (CP) of wheat samples

	Source	Variety or sample ID	MC (%)	IVV (mPa-s)	SW (kg/hl)	GE (MJ/kg DM)	CP (g/kg DM
1	Northern Ireland	Duxford	23.0	9.88	65.0	17.7	101.5
2	Northern Ireland	Grafton	21.0	13.6	72.2	18.0	115.5
3	Northern Ireland	Alchemy	21.9	9.51	70.9	17.8	112.4
4	Republic of Ireland	1		9.64	67.8	17.7	112.9
5	USA			6.86	79.9	17.6	110.8
6	Republic of Ireland	2		8.15	74.2	17.9	117.6
7	France			8.27	75.8	17.9	113.9
8	Germany	SA358		8.28	82.2	18.0	128.4
9	Great Britain	SA359		8.81	76.5	17.9	105.9
10	Great Britain	Solstice	17.3	9.23	70.8	18.0	126.9
11	Great Britain	Brockton	18.5	10.5	63.9	17.8	110.6
12	Great Britain	Neil Furniss		11.4	55.7	17.8	109.6
13	Great Britain	Crusoe	15.5	9.65	75.1	18.0	121.4
14	Great Britain	Myriad	14.9	11.2	70.0	17.7	102.5
15	Great Britain	Alchemy	16.0	10.6	70.1	17.8	116.9
16	Great Britain	Aviator	13.5	8.71	73.6	17.5	90.2
17	Northern Ireland	Composite 1		8.32	58.8	17.8	118.6
18	Northern Ireland	Composite 2		8.65	58.9	17.9	115.4
19	Northern Ireland	Composite 4		11.4	66.4	17.8	117.1
20	Northern Ireland	Composite 5		11.0	69.2	17.8	114.0

Table 6. Starch, microdochium, DON and ZON contents of wheat samples

	Source	Variety or sample ID	Starch (g/kg DM)	Microdochium (pg/ng)	DON (ng/g)	ZON (ng/g)
1	Northern Ireland	Duxford	609.4	15.5	129.9	38.4
2	Northern Ireland	Grafton	626.8	38.1	64.09	N/D
3	Northern Ireland	Alchemy	619.9	40.5	111.3	6.82
4	Republic of Ireland	1	613.4	23.9	85.71	1.94
5	USA		629.9	0.45	363.1	N/D
6	Republic of Ireland	2	618.6	3.10	22.02	N/D
7	France		624.5	6.59	412.8	4.89
8	Germany	SA358	589.2	1.56	71.91	N/D
9	Great Britain	SA359	631.1	8.50	223.6	N/D
10	Great Britain	Solstice	571.0	25.8	314.5	N/D
11	Great Britain	Brockton	640.7	27.6	113.4	N/D
12	Great Britain	Neil Furniss	610.7	18.9	394.5	7.18
13	Great Britain	Crusoe	611.2	41.1	623.4	25.1
14	Great Britain	Myriad	624.4	7.88	142.8	4.13
15	Great Britain	Alchemy	610.5	30.2	677.8	N/D
16	Great Britain	Aviator	667.9	1.04	115.7	N/D
17	Northern Ireland	Composite 1	567.2	38.1	150.6	9.33
18	Northern Ireland	Composite 2	600.0	60.7	180.2	N/D
19	Northern Ireland	Composite 4	605.0	34.9	86.1	N/D
20	Northern Ireland	Composite 5	598.4	25.7	77.1	3.26

N/D = Not detected. Ochratoxin A detected in samples 1, 3 and 8 at levels of 10.57, 0.43 and 0.53 ng/g, respectively. Ochratoxin B detected in sample 1 at a level of 1.71 ng/g.

Table 7. Amino acid content (%) of wheat samples

	Source	Variety or sample ID	Trp	Ala	Arg	Asp	Cys	Glu	Gly	His	Ile	Leu
1	Northern Ireland	Duxford	0.08	0.41	0.49	0.56	0.21	2.71	0.44	0.24	0.37	0.65
2	Northern Ireland	Grafton	0.06	0.36	0.43	0.51	0.19	2.64	0.40	0.21	0.37	0.67
3	Northern Ireland	Alchemy	0.10	0.40	0.50	0.55	0.21	2.91	0.43	0.23	0.41	0.70
4	Republic of Ireland	1	0.09	0.41	0.50	0.56	0.21	3.13	0.45	0.24	0.40	0.71
5	USA		0.11	0.40	0.51	0.57	0.24	3.04	0.45	0.25	0.41	0.71
6	Republic of Ireland	2	0.10	0.42	0.50	0.55	0.22	3.12	0.48	0.25	0.41	0.75
7	France		0.11	0.40	0.51	0.57	0.22	3.08	0.45	0.23	0.41	0.71
8	Germany	SA358	0.13	0.43	0.57	0.62	0.25	3.79	0.51	0.28	0.45	0.80
9	Great Britain	SA359	0.12	0.40	0.49	0.55	0.21	3.02	0.44	0.23	0.37	0.67
10	Great Britain	Solstice	0.09	0.45	0.59	0.65	0.23	3.43	0.52	0.27	0.44	0.78
11	Great Britain	Brockton	0.11	0.40	0.50	0.57	0.21	3.02	0.44	0.23	0.39	0.70
12	Great Britain	Neil Furniss	0.10	0.41	0.50	0.60	0.21	2.85	0.44	0.23	0.37	0.68
13	Great Britain	Crusoe	0.12	0.41	0.53	0.58	0.22	3.35	0.49	0.25	0.42	0.76
14	Great Britain	Myriad	0.11	0.45	0.59	0.65	0.29	3.54	0.50	0.26	0.42	0.76
15	Great Britain	Alchemy	0.10	0.42	0.5	0.59	0.24	3.28	0.45	0.26	0.43	0.73
16	Great Britain	Aviator	0.09	0.35	0.42	0.54	0.24	2.75	0.39	0.21	0.36	0.63
17	Northern Ireland	Composite 1	0.09	0.47	0.56	0.70	0.27	3.68	0.51	0.30	0.48	0.82
18	Northern Ireland	Composite 2	0.10	0.42	0.54	0.6	0.21	3.12	0.45	0.24	0.38	0.73
19	Northern Ireland	Composite 4	0.11	0.52	0.61	0.74	0.32	4.18	0.56	0.33	0.51	0.91
20	Northern Ireland	Composite 5	0.10	0.42	0.53	0.61	0.24	3.36	0.47	0.26	0.42	0.75

Table 7. cont. Amino acid content (%) of wheat samples

	Source	Variety or sample ID	Lys	Met	Phe	Pro	Ser	Thr	Tyr	Val
1	Northern Ireland	Duxford	0.31	0.16	0.43	0.73	0.35	0.27	0.09	0.50
2	Northern Ireland	Grafton	0.27	0.15	0.38	0.96	0.33	0.24	0.07	0.43
3	Northern Ireland	Alchemy	0.32	0.16	0.45	1.04	0.35	0.26	0.10	0.50
4	Republic of Ireland	1	0.31	0.16	0.46	1.17	0.38	0.27	0.11	0.50
5	USA		0.31	*	0.37	1.28	0.36	0.27	0.04	0.49
6	Republic of Ireland	2	0.33	0.17	0.46	1.16	0.38	0.28	0.10	0.51
7	France		0.32	0.17	0.44	1.34	0.37	0.27	0.08	0.49
8	Germany	SA358	0.33	0.18	0.50	1.41	0.43	0.31	0.09	0.57
9	Great Britain	SA359	0.30	0.16	0.40	1.12	0.38	0.27	0.06	0.48
10	Great Britain	Solstice	0.35	0.17	0.48	1.12	0.43	0.31	0.12	0.55
11	Great Britain	Brockton	0.30	0.16	0.43	0.92	0.39	0.28	0.07	0.48
12	Great Britain	Neil Furniss	0.32	0.16	0.42	0.86	0.38	0.28	0.10	0.50
13	Great Britain	Crusoe	0.32	0.16	0.46	0.96	0.45	0.30	0.07	0.51
14	Great Britain	Myriad	0.38	0.21	0.43	1.08	0.49	0.33	0.13	0.55
15	Great Britain	Alchemy	0.30	0.17	0.47	1.15	0.42	0.28	0.10	0.52
16	Great Britain	Aviator	0.30	0.16	0.35	1.11	0.38	0.25	0.09	0.44
17	Northern Ireland	Composite 1	0.35	0.19	0.49	1.22	0.48	0.32	0.11	0.59
18	Northern Ireland	Composite 2	0.32	0.17	0.49	1.02	0.5	0.32	0.14	0.47
19	Northern Ireland	Composite 4	0.38	0.21	0.56	0.95	0.52	0.36	0.11	0.64
20	Northern Ireland	Composite 5	0.32	0.18	0.41	0.90	0.43	0.30	0.09	0.54

Table 8. Minimum, mean, maximum and standard deviation values of the amino acid content (%) of the wheat samples

	Minimum	Mean	Maximum	Standard Deviation
Tryptophan	0.6	1.0	1.3	0.16
Alanine	3.5	4.2	5.2	0.36
Arginine	4.2	5.2	6.1	0.49
Aspartic acid	5.1	5.9	7.4	0.56
Cysteine	1.9	2.3	3.2	0.31
Glutamic acid	26.4	32.0	41.8	3.87
Glycine	3.9	4.6	5.6	0.42
Histidine	2.1	2.5	3.3	0.29
Isoleucine	3.6	4.1	5.1	0.39
Leucine	6.3	7.3	9.1	0.65
Lysine	2.7	3.2	3.8	0.27
Methionine	1.5	1.7	2.1	0.17
Phenylalanine	3.5	4.4	5.6	0.49
Proline	7.3	10.8	14.1	1.68
Serine	3.3	4.1	5.2	0.55
Threonine	2.4	2.9	3.6	0.30
Tyrosine	0.4	0.9	1.4	0.24
Valine	4.3	5.1	6.4	0.50

Table 9. The relationship (R^2) between wheat chemical and physical parameters

	MC	IVV	SW	GE	CP	Starch	Microdochium	DON	ZON
MC									
IVV	0.08 (0.462)								
SW	0.13 (0.186)	0.07 (0.131)							
GE	0.05 (0.558)	0.01 (0.712)	0.02 (0.544)						
CP	0.02 (0.720)	0.00 (0.791)	0.01 (0.612)	**0.55 (<0.001)**					
Starch	0.06 (0.513)	0.00 (0.807)	0.03 (0.224)	0.14 (0.059)	**0.53 (<0.001)**				
Microdochium	0.06 (0.258)	0.08 (0.117)	**0.29 (0.009)**	0.05 (0.177)	0.07 (0.145)	0.10 (0.093)			
DON	0.06 (0.258)	0.01 (0.706)	0.01 (0.736)	0.03 (0.456)	0.03 (0.450)	0.00 (0.818)	0.09 (0.697)		
ZON	0.16 (0.597)	0.01 (0.813)	0.00 (1.000)	0.05 (0.571)	0.04 (0.608)	0.003 (0.885)	0.01 (0.786)	0.06 (0.515)	

Probability values are in parenthesis. Significant relationships ($P<0.05$) are in bold

4.2. AFBI performance trial

The effect of wheat sample on broiler performance is shown in Table 10. There was no significant (P>0.05) effect on performance in the starter or grower phase. However, there were significant differences due to wheat sample in the finishing stage. DMI ranged from 1564g (wheat 8, SA358, Germany to 1869g (wheat 5, USA) (SEM = 62.3, P=0.036). LWG ranged from 1093g (wheat 17, Composite 1, NI) to 1378g (wheat 2, Grafton, NI) (SEM=48.0, P=0.028). FCR ranged from 1.31 (wheat 19, Composite 4, NI) to 1.54 (wheat 5, USA) (SEM=0.035, P<0.001). Wheat sample also had a significant effect on body weight at 21, 28 and 35d. Body weight at 21d was lowest for birds offered the diet based on wheat 16, Avator, GB (930g) and highest for birds offered the diet based on wheat 1, Duxford, NI (1039g) (SEM=19.7, P=0.041). The diet based on wheat 16, Avator, GB also resulted in the lowest liveweight at 28d (1483g) and the second lowest at 35d (2061g). Birds offered the diet based on wheat 2, Grafton, NI had the highest liveweight at 28d (1675g) and 35d (2371g). Those offered the diet based on wheat 1, Duxford, NI had the second highest liveweight at 35d (2236g).

Table 11 shows the effect of wheat moisture content at harvest on broiler performance. There were significant linear effects on LWG in the starter stage (P<0.001), the finisher stage (P=0.044) and also over the overall period (P=0.005). As wheat moisture content at harvest increased, LWG increased in the starter period (R^2=0.57, P<0.001), finisher period (R^2=0.23, P=0.044) and over the overall period (R^2=0.40, P=0.005). Similarly, as wheat moisture content increased, liveweight was increased at 14d (R^2=0.57, P<0.001), 21d (R^2=0.43, P<0.003), 28d (R^2=0.31, P=0.003) and 35d (R^2=0.40, P<0.005). In the starter period, increasing wheat moisture content at harvest improved FCR (R^2=0.75, P=0.01).

Wheat microdochium content had a significant linear effect on DMI in the grower stage (P=0.047), on finisher FCR (P=0.006) and on overall FCR (P=0.048) (Table 12). However, while significant, the relationships were not strong (R^2 = 0.16, 0.17 and 0.19, respectively). In order to investigate these effects further, historic wheat samples were analysed for microdochium content and results correlated with recorded performance (Owens *et al* 2007). The minimum, mean, maximum and standard deviation values of these historic wheats are shown in Table 13. When correlated with DMI, LWG and FCR, similar strength relationships were observed (R^2=0.22, 0.18 and 0.10, respectively).

Wheat DON content had a significant effect (P<0.05) on broiler performance in the finisher stage (Table 14). DON content also significantly affected liveweight at 21d, 28d and 35d. There was no significant linear relationship between wheat DON content and bird performance. However, there was a quadratic relationship between DON content and finisher FCR (P=0.002), overall DMI (P=0.026) and overall FCR (P=0.032). There were significant effects of wheat ZON content on finisher DMI (0.02) and 35d liveweight (P=0.034). DON content also had significant linear effects on grower DMI (R^2=0.41, P=0.017), overall DMI (R^2=0.61, P=0.004) and overall LWG (R^2=0.26, P=0.032), 14d liveweight (R^2= =0.49, P=0.016) and 35d liveweight (R^2=0.27, P=0.033) (Table 15). There were also significant quadratic effects observed for grower FCR (P=0.023) and finisher DMI (P=0.022).

There were no strong relationships between wheat chemical composition and broiler performance (Table 16). However, *in vitro* viscosity was found to be negatively related (P<0.05) to LWG and FCR (R^2=0.22 and 0.39, respectively).

Table 10. The effect of wheat sample on broiler performance (AFBI trial)

	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Starter DMI (g)	484	468	471	473	488	468	454	459	483	450	459	491	482	497
Starter LWG (g)	478	447	448	453	458	449	431	427	437	417	436	443	442	440
Starter FCR	1.02	1.05	1.05	1.04	1.07	1.05	1.05	1.07	1.12	1.09	1.05	1.11	1.10	1.13
Grower DMI (g)	695	667	690	652	672	672	642	642	631	639	681	695	713	671
Grower LWG (g)	517	502	511	505	510	497	498	484	497	494	522	528	494	530
Grower FCR	1.35	1.33	1.35	1.29	1.32	1.35	1.29	1.33	1.28	1.30	1.31	1.32	1.45	1.28
Finisher DMI (g)	1813	1816	1643	1610	1869	1641	1610	1564	1713	1706	1697	1717	1645	1638
Finisher LWG (g)	1297	1378	1211	1185	1232	1179	1142	1159	1251	1252	1163	1274	1228	1215
Finisher FCR	1.40	1.32	1.36	1.36	1.54	1.39	1.41	1.35	1.37	1.37	1.43	1.35	1.34	1.35
Overall DMI (g)	3350	3172	3083	3033	3401	3031	3007	3000	3156	3104	3167	3216	3111	3097
Overall LWG (g)	2291	2327	2170	2144	2201	2124	2070	2070	2184	2163	2121	2244	2163	2185
Overall FCR	1.46	1.37	1.42	1.42	1.56	1.43	1.45	1.45	1.45	1.44	1.49	1.43	1.44	1.42
7d wt (g)	178	161	170	171	174	168	164	164	169	158	166	171	168	164
14d wt (g)	522	491	493	498	502	493	475	471	481	461	480	487	486	484
21d wt (g)	1039	993	1004	1003	1012	989	973	955	978	955	1002	1014	980	1015
28d wt (g)	1669	1675	1597	1577	1614	1561	1584	1515	1588	1560	1586	1620	1571	1608
35d wt (g)	2336	2371	2215	2188	2245	2169	2115	2114	2228	2208	2166	2288	2207	2229

Table 10. cont. The effect of wheat sample on broiler performance (AFBI trial)

	15	16	17	18	19	20	SEM	P
Starter DMI	458	460	471	484	438	469	17.5	0.745
Starter LWG	437	410	434	440	429	432	11.8	0.095
Starter FCR	1.05	1.12	1.09	1.11	1.02	1.09	0.036	0.581
Grower DMI	668	648	683	672	661	641	19.2	0.172
Grower LWG	502	475	490	523	495	486	13.7	0.256
Grower FCR	1.33	1.37	1.40	1.29	1.34	1.33	0.047	0.689
Finisher DMI	1702	1623	1596	1694	1559	1640	62.3	0.036
Finisher LWG	1302	1150	1093	1264	1197	1223	48.0	0.024
Finisher FCR	1.32	1.42	1.46	1.34	1.31	1.34	0.035	<0.001
Overall DMI	3077	3078	3042	3134	3006	3025	83.5	0.052
Overall LWG	2241	2036	2017	2227	2121	2142	55.4	0.1
Overall FCR	1.38	1.52	1.51	1.41	1.42	1.41	0.045	0.34
7d wt (g)	167	161	166	166	163	160	5.2	0.593
14d wt (g)	482	455	478	484	473	477	11.8	0.096
21d wt (g)	984	930	968	1007	969	963	19.7	0.041
28d wt (g)	1645	1483	1525	1614	1539	1581	37.1	0.047
35d wt (g)	2286	2080	2061	2271	2165	2186	55.4	0.01

Table 11. The effect of wheat moisture content at harvest on broiler performance

	13.5%	14.9%	15.5%	16%	17.3%	18.5%	21%	21.9%	23%	SEM	P	P=Lin	P=Quad
Starter DMI (g)	460	497	482	458	450	459	468	471	484	17.47	0.636	0.848	0.318
Starter LWG (g)	410	440	442	437	417	436	447	448	478	11.78	0.741	<0.001	0.336
Starter FCR	1.12	1.13	1.10	1.05	1.09	1.05	1.05	1.05	1.02	0.036	0.72	0.010	0.700
Grower DMI (g)	448	671	713	668	639	681	667	690	695	19.15	0.246	0.233	0.809
Grower LWG (g)	475	530	494	502	494	522	502	511	517	13.7	0.44	0.169	0.442
Grower FCR	1.37	1.28	1.45	1.33	1.30	1.31	1.33	1.35	1.35	0.047	0.838	0.863	0.355
Finisher DMI (g)	1623	1638	1645	1702	1706	1697	1816	1643	1813	62.3	0.028	0.224	0.024
Finisher LWG (g)	1150	1215	1228	1302	1252	1163	1378	1211	1297	48	0.109	0.044	0.496
Finisher FCR	1.42	1.35	1.34	1.32	1.37	1.46	1.32	1.36	1.40	0.035	0.079	0.963	0.616
Overall DMI (g)	3078	3097	3111	3077	3104	3167	3172	3083	3350	83.5	0.376	0.050	0.422
Overall LWG (g)	2036	2185	2163	2241	2163	2121	2327	2170	2291	55.4	0.062	0.005	0.565
Overall FCR	1.52	1.42	1.44	1.38	1.44	1.50	1.37	1.42	1.46	0.047	0.302	0.402	0.276
7d wt (g)	161	164	168	167	158	166	161	170	178	5.16	0.256	0.085	0.200
14d wt (g)	455	484	486	482	461	480	491	493	522	11.79	0.011	<0.001	0.331
21d wt (g)	930	1015	980	984	955	1002	993	1004	1039	19.74	0.011	0.003	0.962
28d wt (g)	1483	1608	1571	1645	1560	1586	1675	1597	1669	37.13	0.012	0.003	0.393
35d wt (g)	2080	2229	2207	2286	2208	2166	2371	2215	2336	55.4	0.016	0.005	0.567

Table 12. The effect of wheat microdochium (pg/ng) content on broiler performance

	0.45	1.04	1.56	3.10	6.59	7.88	8.50	15.52	18.92	23.91	25.70	25.82	27.62	30.23	34.92
Starter DMI (g)	488	460	459	468	454	497	483	484	491	473	469	450	459	458	438
Starter LWG (g)	458	410	427	449	431	440	437	478	443	453	432	417	436	437	429
Starter FCR	1.07	1.12	1.07	1.05	1.05	1.13	1.12	1.02	1.11	1.04	1.09	1.09	1.05	1.05	1.02
Grower DMI (g)	672	648	642	672	642	671	631	695	695	652	641	639	681	668	661
Grower LWG (g)	510	475	484	497	498	530	497	517	528	505	486	494	522	502	495
Grower FCR	1.32	1.37	1.33	1.35	1.29	1.28	1.28	1.35	1.32	1.29	1.33	1.30	1.31	1.33	1.34
Finisher DMI (g)	1869	1623	1564	1641	1610	1638	1713	1813	1717	1610	1640	1706	1697	1702	1559
Finisher LWG (g)	1232	1150	1159	1179	1142	1215	1251	1297	1274	1185	1223	1252	1163	1302	1197
Finisher FCR	1.54	1.42	1.35	1.39	1.41	1.35	1.37	1.40	1.35	1.36	1.34	1.37	1.43	1.32	1.31
Overall DMI (g)	3401	3078	3000	3031	3007	3097	3156	3350	3216	3033	3025	3104	3167	3077	3006
Overall LWG (g)	2201	2036	2070	2124	2070	2185	2184	2291	2244	2144	2142	2163	2121	2241	2121
Overall FCR	1.56	1.52	1.45	1.43	1.45	1.42	1.45	1.46	1.43	1.42	1.41	1.44	1.49	1.38	1.42
7d wt (g)	174	161	164	168	164	164	169	178	171	171	160	158	166	167	163
14d wt (g)	502	455	471	493	475	484	481	522	487	498	477	461	480	482	473
21d wt (g)	1012	930	955	989	973	1015	978	1039	1014	1003	963	955	1002	984	969
28d wt (g)	1614	1483	1515	1561	1584	1608	1588	1669	1620	1577	1581	1560	1586	1645	1539
35d wt (g)	2245	2080	2114	2169	2115	2229	2228	2336	2288	2188	2186	2208	2166	2286	2165

Table 12 cont. The effect of microdochium content on broiler performance

	38.09	38.10	40.48	41.06	60.66	SEM	P	P=Lin	P=Quad	R^2
Starter DMI (g)	468	471	471	482	484	17.5	0.745	0.843	0.369	
Starter LWG (g)	447	434	448	442	440	11.8	0.095	0.843	0.735	
Starter FCR	1.05	1.09	1.05	1.10	1.11	0.036	0.581	0.682	0.201	
Grower DMI (g)	667	683	690	713	672	19.2	0.172	**0.047**	0.758	0.16
Grower LWG (g)	502	490	511	494	523	13.7	0.256	0.323	0.872	
Grower FCR	1.33	1.40	1.35	1.45	1.29	0.047	0.689	0.442	0.724	
Finisher DMI (g)	1816	1596	1643	1645	1694	62.3	**0.036**	0.842	0.887	
Finisher LWG (g)	1378	1093	1211	1228	1264	48.0	**0.024**	0.106	0.548	
Finisher FCR	1.32	1.46	1.36	1.34	1.34	0.035	**<0.001**	**0.006**	0.229	0.17
Overall DMI (g)	3172	3042	3083	3111	3134	83.5	0.052	0.51	0.668	
Overall LWG (g)	2327	2017	2170	2163	2227	55.4	0.100	0.092	0.581	
Overall FCR	1.37	1.51	1.42	1.44	1.41	0.045	0.340	**0.048**	0.387	0.19
7d wt (g)	161	166	170	168	166	5.2	0.593	0.644	0.945	
14d wt (g)	491	478	493	486	484	11.8	0.096	0.832	0.740	
21d wt (g)	993	968	1004	980	1007	19.7	**0.041**	0.416	0.932	
28d wt (g)	1675	1525	1597	1571	1614	37.1	**0.047**	0.169	0.393	
35d wt (g)	2371	2061	2215	2207	2271	55.4	**0.01**	0.092	0.582	

Table 13. Microdochium content (pg/ng) of historic wheat samples (n=59) used in the study of Owens *et al* (2007) and of those used in the current study

	Owens *et al* (2007) (n=59)	Current study (n=20)
Minimum	0.002	0.45
Mean	1.06	22.51
Maximum	5.69	60.77
Standard Deviation	1.48	16.80

Table 14. The effect of DON (ng/g) content on broiler performance

	22.02	**64.09**	**71.91**	**77.06**	**85.71**	**86.14**	**111.29**	**113.35**	**115.69**	**129.85**	**142.78**	**150.63**	**180.21**
Starter DMI (g)	468	468	459	469	473	438	471	459	460	484	497	471	484
Starter LWG (g)	449	447	427	432	453	429	448	436	410	478	440	434	440
Starter FCR	1.05	1.05	1.07	1.09	1.04	1.02	1.05	1.05	1.12	1.02	1.13	1.09	1.11
Grower DMI (g)	672	667	642	641	652	661	690	681	648	695	671	683	672
Grower LWG (g)	497	502	484	486	505	495	511	522	475	517	530	490	523
Grower FCR	1.35	1.33	1.33	1.33	1.29	1.34	1.35	1.31	1.37	1.35	1.28	1.40	1.29
Finisher DMI (g)	1641	1816	1564	1640	1610	1559	1643	1697	1623	1813	1638	1596	1694
Finisher LWG (g)	1179	1378	1159	1223	1185	1197	1211	1163	1150	1297	1215	1093	1264
Finisher FCR	1.39	1.32	1.35	1.34	1.36	1.31	1.36	1.43	1.42	1.40	1.35	1.46	1.34
Overall DMI (g)	3031	3172	3000	3025	3033	3006	3083	3167	3078	3350	3097	3042	3134
Overall LWG (g)	2124	2327	2070	2142	2144	2121	2170	2121	2036	2291	2185	2017	2227
Overall FCR	1.43	1.37	1.45	1.41	1.42	1.42	1.42	1.49	1.52	1.46	1.42	1.51	1.41
7d wt (g)	168	161	164	160	171	163	170	166	161	178	164	166	166
14d wt (g)	493	491	471	477	498	473	493	480	455	522	484	478	484
21d wt (g)	989	993	955	963	1003	969	1004	1002	930	1039	1015	968	1007
28d wt (g)	1561	1675	1515	1581	1577	1539	1597	1586	1483	1669	1608	1525	1614
35d wt (g)	2169	2371	2114	2186	2188	2165	2215	2166	2080	2336	2229	2061	2271

Table 14 cont. The effect of DON (ng/g) on broiler performance

	223.56	**314.45**	**363.07**	**394.52**	**412.77**	**623.39**	**677.84**	SEM	P	P=Lin	P=Quad
Starter DMI (g)	483	450	488	491	454	482	458	17.5	0.745	0.709	0.312
Starter LWG (g)	437	417	458	443	431	442	437	11.8	0.095	0.833	0.758
Starter FCR	1.12	1.09	1.07	1.11	1.05	1.10	1.05	0.036	0.581	0.478	0.135
Grower DMI (g)	631	639	672	695	642	713	668	19.2	0.172	0.195	0.404
Grower LWG (g)	497	494	510	528	498	494	502	13.7	0.256	0.743	0.178
Grower FCR	1.28	1.30	1.32	1.32	1.29	1.45	1.33	0.047	0.689	0.488	0.086
Finisher DMI (g)	1713	1706	1869	1717	1610	1645	1702	62.3	0.036	0.255	0.133
Finisher LWG (g)	1251	1252	1232	1274	1142	1228	1302	48.0	0.024	0.166	0.571
Finisher FCR	1.37	1.37	1.54	1.35	1.41	1.34	1.32	0.035	<0.001	0.739	0.002
Overall DMI (g)	3156	3104	3401	3216	3007	3111	3077	83.5	0.052	0.324	0.026
Overall LWG (g)	2184	2163	2201	2244	2070	2163	2241	55.4	0.100	0.216	0.825
Overall FCR	1.45	1.44	1.56	1.43	1.45	1.44	1.38	0.045	0.340	0.982	0.032
7d wt (g)	169	158	174	171	164	168	167	5.2	0.593	0.505	0.707
14d wt (g)	481	461	502	487	475	486	482	11.8	0.096	0.822	0.739
21d wt (g)	978	955	1012	1014	973	980	984	19.7	0.041	0.926	0.429
28d wt (g)	1588	1560	1614	1620	1584	1571	1645	37.1	0.047	0.172	0.98
35d wt (g)	2228	2208	2245	2288	2115	2207	2286	55.4	0.01	0.218	0.821

Table 15. The effect of ZON (ng/g) on broiler performance

	1.94	**3.26**	**4.13**	**4.89**	**6.82**	**7.18**	**9.33**	**25.07**	**38.4**	SEM	P	P=Lin	R^2	P=Quad
Starter DMI (g)	473	469	497	454	471	491	471	482	484	17.5	0.806	0.568		0.964
Starter LWG (g)	453	432	440	431	448	443	434	442	478	11.8	0.165	0.016		0.078
Starter FCR	1.04	1.09	1.13	1.05	1.05	1.11	1.09	1.10	1.02	0.036	0.382	0.253		0.143
Grower DMI (g)	652	641	671	642	690	695	683	713	695	19.2	0.085	0.017	0.41	0.036
Grower LWG (g)	505	486	530	498	511	528	490	494	517	13.7	0.22	0.829		0.398
Grower FCR	1.29	1.33	1.28	1.29	1.36	1.31	1.40	1.45	1.35	0.047	0.209	0.068		0.023
Finisher DMI (g)	1610	1640	1638	1610	1643	1717	1596	1645	1813	62	0.02	0.307		0.022
Finisher LWG (g)	1185	1223	1215	1142	1211	1274	1093	1228	1297	48	0.111	0.068		0.0292
Finisher FCR	1.36	1.34	1.35	1.41	1.36	1.35	1.46	1.34	1.40	0.035	0.189	0.737		0.91
Overall DMI (g)	3033	3025	3097	3007	3083	3216	3042	3111	3350	83.5	0.095	0.004	0.61	0.405
Overall LWG (g)	2144	2142	2185	2070	2170	2244	2017	2163	2291	55.4	0.034	0.032	0.26	0.136
Overall FCR	1.42	1.41	1.42	1.45	1.42	1.43	1.51	1.44	1.46	0.045	0.847	0.501		0.513
7d wt (g)	171	160	164	164	170	171	166	168	178	5.2	0.409	0.052		0.544
14d wt (g)	498	477	484	475	493	487	478	486	522	11.8	0.166	0.016	0.49	0.079
21d wt (g)	1003	963	1015	973	1004	1014	968	980	1039	19.7	0.110	0.110		0.103
28d wt (g)	1577	1581	1608	1584	1597	1619.7	1525	1571.2	1669	37.1	0.362	0.132		0.103
35d wt (g)	2188	2186	2229	2115	2215	2288	2061	2207	2336	55.4	0.034	0.033	0.27	0.136

Table 16. The relationship (R^2) between chemical composition and overall broiler performance (0-35d)

	IVV	SW	GE	CP	Starch
DMI	0.00 (0.833)	0.01 (0.763)	0.01 (0.310)	0.08 (0.123)	0.00 (0.314)
LWG	0.22 **(0.020)**	0.03 (0.471)	0.02 (0.585)	0.01 (0.740)	0.00 (0.807)
FCR	0.39 **(0.002)**	0.02 (0.562)	0.13 (0.062)	0.03 (0.215)	0.04 (0.410)

Probability values are in parenthesis. Significant relationships are in bold

4.3. Chemical and physical analysis of wheat used in the Moy Park trial

Table 17 shows specific weight values and the analysed level of moisture content, oil A, crude protein, fibre, ash and starch in the wheat samples used in the trial. Oil A ranged from 10.3 to 13.3 g/kg, crude protein ranged from 81.3 to 115.6 g/kg, fibre ranged from 22.4 to 26.7g/kg, ash ranged from 11.4 to 13.8g/kg and starch content ranged from 540.0 to 564.8g/kg.

Table17. Chemical composition (g/kg fresh) and specific weight (SW) (kg/hL) of wheat samples used in Moy Park trial

	Moisture (%)	SW	Oil A	Protein	Fibre	Ash	Starch
Danish Old Crop	15.4	77.2	10.3	93.2	22.4	11.4	557.0
Romanian New Crop	14.8	78.4	12.0	115.6	22.6	13.8	564.8
UK Old Crop	14.9	74.4	11.7	103.3	26.0	12.3	548.3
UK New Crop	13.9	77.5	13.3	95.7	23.7	12.3	563.7
Irish New Crop	14.9	76.3	12.3	104.0	26.7	13.0	540.0
Danish New Crop	13.6	77.8	15.1	81.3	24.0	12.2	N/D

N/D = Not determined

4.4. Moy Park performance trial

The different wheat samples used to formulate broiler diets had no significant effect on bird performance (Table 18). However, the numerical differences between overall intake and FCR were substantial (3323 to 3767g and 1.48 to 1.67, respectively). There was a trend (P=0.074) for bird weight at 7d of age to be slightly heavier for birds offered diets based on Irish new crop wheat (168g) than those offered diets based on UK old crop wheat (159g). The origin of wheat did not significantly affect bird performance (Table 19). Although, again, there was a tendency (P=0.058) for Irish wheat to result in slightly heavier birds at 7d of age (168g) compared with birds offered diets based on UK wheat (161g). Table 20 presents the effect of offering diets based on new crop or old crop wheat. The only significant (P=0.026) effect was observed on bird weight at 7d of age, with those offered new crop wheat being slightly heavier (165g) than those offered diets based on old crop wheat (161g, SEM=1.4).

Table 18. The effect of wheat sample on broiler performance

	Danish old	Romanian new	UK old	UK new	Irish new	Danish new	SEM	P
Overall intake (g)	3648	3649	3767	3323	3577	3412	140.6	0.347
Overall LWG (g)	2249	2204	2223	2246	2214	2221	27.5	0.829
Overall FCR	1.62	1.66	1.67	1.48	1.62	1.54	0.066	0.305
7d wt (g)	163	164	159	162	168	166	2.2	0.074
14d wt (g)	445	433	430	443	444	440	5.8	0.386
21d wt (g)	911	904	888	934	917	916	12.1	0.185
28d wt (g)	1501	1493	1527	1547	1506	1528	20.3	0.434
38d wt (g)	2294	2249	2267	2291	2259	2265	27.6	0.83

Table 19. The effect of wheat origin on broiler performance

	Danish	Irish	Romanian	UK	SEM	SEM	P
Overall intake (g)	3530	3577	3649	3515	147.2	104.1	0.887
Overall LWG (g)	2235	2214	2204	2234	27	19.1	0.741
Overall FCR	1.58	1.62	1.66	1.57	0.07	0.049	0.752
7d wt (g)	164	168	164	161	2.2	1.6	0.058
14d wt (g)	443	444	433	436	5.9	4.2	0.412
21d wt (g)	914	917	904	911	13.1	9.3	0.897
28d wt (g)	1515	1506	1493	1537	20	14.2	0.308
38d wt (g)	2279	2259	2249	2279	27.1	19.2	0.746

Table 20. The effect of old or new crop wheat on broiler performance

	New crop	Old crop	SEM	SEM	P
Overall intake (g)	3490	3677	98.6	69.7	0.13
Overall LWG (g)	2222	2236	18.8	13.3	0.536
Overall FCR	1.57	1.65	0.047	0.034	0.211
7d wt (g)	165	161	1.6	1.1	0.026
14d wt (g)	440	438	4.2	3	0.713
21d wt (g)	918	900	8.7	6.2	0.096
28d wt (g)	1519	1514	14.5	10.3	0.809
38d wt (g)	2266	2280	18.8	13.3	0.541

4.5. Harper Adams performance trial

See Appendix 3 for the full report.

4.6. NIRS prediction equations

A glossary of terms used to assist in the interpretation of NIRS statistics are presented in Table 21.

Table 21. Definitions of terms used in NIRS

Term	Definition
N	Number of observations used in final calibration
Mean	Mean of experiential observations
SD	Standard deviation
R^2	Fraction of variance accounted for by the NIR calibration when all accepted observations are included in the relationship (i.e. relationship between actual and predicted)
SEC	Standard error of calibration when all accepted observations are included in the relationship
1-VR	1-Variance ratio – Fraction of variance accounted for NIR prediction when some observations are used for cross validation of the calibration
SECV	Standard error of cross validation when some observations are used for cross validation of the calibration
SECV as % of the mean	Indication of accuracy of calibration with values of less than 5% being acceptable
RPD calibration (Black 2008)	Ratio of prediction to deviation = SD/SECV RPD<1.5 = calibration unsatisfactory RPD 1.5-2.0 = calibration can distinguish between high and low values RPD 2.0-2.5 = calibration quantitative RPD 2.5-3.0 = calibration predictions good RPD >3.0 = calibration predictions excellent
SEP	Standard error of prediction when some values are used for independent validation
SEP as % of the mean	Indication of accuracy of prediction with values of less than 5% being acceptable
RPD prediction	Ratio of prediction to deviation = SD/SEP RPD<1.5 = calibration unsatisfactory RPD 1.5-2.0 = calibration can distinguish between high and low values RPD 2.0-2.5 = calibration quantitative RPD 2.5-3.0 = calibration predictions good RPD >3.0 = calibration predictions excellent

4.6.1 NIRS prediction of wheat microdochium content

The calibration and validation statistics for wheat microdochium content are presented in Table 22. The calibration for the medium content dataset was the strongest (R^2 = 0.94, SEC=0.417) but decreased upon validation (R^2=0.777, SECV=0.765 and SECV as % of mean=14%). The prediction equation using the entire dataset resulted in a reasonably robust 1-VR (0.70). However, the SECV as % of mean was extremely high (109%) and the RPD unacceptable. The independent cross validation statistics showing the strength of relationship between actual and predicted wheat microdochium content are presented in Table 23. When the entire dataset was considered, the R^2 was 0.8 but the SEP and SEP as % of the mean were high (4.972 and 164%) reflective of the fact that the dataset was unevenly spread. When the dataset was split into low and medium categories SEP and SEP as % of the mean were substantially improved (SEP=0.368 and 0.361 and SEP as % of mean=52 and 7%, respectively). Performing cross validation statistics on the high content dataset did not improve SEP (6.968) but did improve SEP as % of mean (22%). RPD was best for the medium dataset at 4.61.

4.6.2 NIRS prediction of broiler performance

The calibration and validation statistics for the prediction of broiler performance are presented in Table 24. Data from all three trials were used to generate these results. The calibration and prediction of feed intake, LWG and feed efficiency (expressed as % gain:feed) were robust with R^2 of 0.92, 0.88 and 0.89, respectively. The SECV were low (3.08, 2.81 and 2.11, respectively) as were the SECV of the mean (3.9, 4.9 and 2.9, respectively) indicating that there is good correlation between actual and predicted values. In addition, RPD for feed intake indicated that the calibration prediction was excellent (4.12). The independent cross validation statistics showing the strength of relationship between actual and predicted wheat microdochium content are presented in Table 25. The relationship between actual and predicted feed intake (R^2=0.92), LWG (R^2=0.83) and gain:feed (%) (R^2=0.85) were strong and SEP low (2.955, 2.787 and 2.023, respectively). The SEP as % of the mean were also low (<5% for all parameters) which indicates that the error in predicting the relationship is acceptable. The best RPD value for feed intake (3.43) and RPD values for LWG and FCR indicated the prediction were quantitative and good, respectively.

In order to test the prediction equations the actual and predicted performance of broilers on the Moy Park trial were compared (Table 26). The relationships (R^2) between actual and predicted feed intake, LWG and feed efficiency were 0.37, 0.46 and 0.46, respectively.

Table 22. The calibration and validation statistics of the prediction of microdochium content

	N	Mean	SD	Min	Max	SEC	RSQ	SECV	1-VR	SECV as % of mean	RPD cal
Entire dataset	142	6.19	12.38	0	43.3	6.231	0.747	6.760	0.700	109	1.83
Low content dataset	100	0.57	0.492	0	2.05	0.367	0.443	0.374	0.417	66	1.32
Medium content dataset	20	5.50	1.663	0.51	10.55	0.417	0.937	0.765	0.777	14	2.17
High content dataset	22	32.44	12.55	0	70.00	7.498	0.643	11.215	0.163	35	1.12

Table 23. Statistics of cross validation for wheat microdochium content (entire data set, n=71, low content data set, n=50, medium content dataset, n=10 and high content dataset, n=11)

	Entire dataset		Low content dataset		Medium content dataset		High content dataset	
	Actual	Predicted	Actual	Predicted	Actual	Predicted	Actual	Predicted
SEP	4.972		0.368		0.361		6.968	
Means	3.025	2.627	0.589	0.563	5.498	5.498	32.35	32.35
R^2	0.804		0.515		0.950		0.677	
SEP as % of mean	164		52		7		22	
Standard deviation	10.93	8.54	0.528	0.345	1.66	1.62	12.55	10.32
RPD validation	2.20		1.43		4.61		1.80	

Table 24. The calibration and validation statistics for the prediction of broiler performance

	N	Mean	SD	Min	Max	SEC	RSQ	SECV	SECV as % of mean	RPD cal
DMI	182	78.46	10.07	48.24	108.77	2.774	0.924	3.083	3.9	4.12
LWG	182	57.30	6.76	37.02	77.66	2.326	0.882	2.806	4.9	2.41
Gain:feed (%)	181	73.16	5.19	57.58	88.73	1.717	0.891	2.107	2.9	2.46

Table 25. Statistics of cross validation for feed intake, LWG and gain:feed (%) (n= 186)

	Feed intake		LWG		Gain:feed (%)	
	Actual	Predicted	Actual	Predicted	Actual	Predicted
SEP	2.955		2.787		2.023	
Means	78.65	78.47	57.32	57.08	73.18	73.13
R^2	0.915		0.832		0.85	
SEP as % of mean	3.8		4.7		2.8	
Standard deviation	10.15	9.70	6.73	6.49	5.23	4.86
RPD validation	3.43		2.41		2.59	

Table 26. Relationship between actual and predicted performance of broilers housed under commercial conditions within the Moy Park trial

	Danish Old Crop	Romanian New Crop	UK Old Crop	UK New Crop	Irish New Crop	Danish New Crop	R^2
Actual intake (g/d)	95.99	96.04	97.56	87.45	94.13	89.80	
Predicted intake (g/d)	93.49	91.34	97.87	91.84	93.32	91.45	0.366
Actual LWG (g/d)	59.19	58.01	58.49	59.11	58.27	58.45	
Predicted LWG (g/d)	56.74	59.96	60.46	58.38	59.89	57.33	0.456
Actual gain:feed (%)*	62.04	60.52	60.46	69.13	62.36	65.41	
Predicted gain:feed (%)*	60.85	63.83	62.03	65.00	63.01	64.38	0.464

*Gain:feed (%) is gain:feed efficiency x 100 and is an output from the prediction statistics

5. Discussion

5.1 Chemical and physical analysis of wheat

The ranges for the measured parameters were within expected limits for wheat (Owens *et al* 2007). However, the aim of sourcing wheat ranging in content of specific parameters was achieved and resulted in wheat samples with differing nutritive values. This allowed for a range of data points when developing the NIRS equations.

The correlations between the majority of the chemical and physical parameters of wheat were not strong and may have been a result of the small dataset (n=20). Starch content was found to be inversely proportional to CP content which is in keeping with the findings of previous research (Ball *et al* 2013a). CP content was also found to be related to energy content, which again is in line with that found previously by Ball *et al* (2013a). However, the strength of the observed relationship in the current study (R^2=0.55) is weaker than that observed by Ball *et al* (2013a) (R^2=0.80) which is reflective of the small dataset in the current study. SW is a measure of the weight of a volume of air-dry grain, expressed in kg/hL. Therefore, low SW grain is a grain of a low density (Millar and Wilkinson 1998). Low values for SW may be due to either immature grain or to grain shape and morphology (Baker *et al* 1965). The latter is influenced by variety while the former is influenced by environmental conditions and time of harvest. Generalisations are made depending on the value of SW. It is assumed that low SW grains contain higher levels of CP, fat and fibre but lower levels of starch (Sibbald and Price 1976). In this study, there were no significant relationships between SW and any of the above parameters. While this finding may be partly due to the small dataset, other workers have reported similar lack of relationships (e.g. Campbell *et al* 1995). The observed lack of relationship between SW and chemical composition again raises questions about the suitability of using SW to predict chemical composition. However, there was a significant relationship between SW and microdochium content, though the correlation was low (R^2=0.29), with microdochium content decreasing as SW value increased. This is not an unexpected finding as microdochium has been shown to reduce grain weight (Eken *et al* 2011).

5.2 Broiler performance studies

In the AFBI and HAU trials, wheat sample significantly affected bird performance. It was been widely established that different samples of wheat result in variable performance and much research has been conducted in the area. Wheat variety, location of production, fertiliser and fungicide addition, storage conditions and new season phenomenon have all been reported to cause variable performance (McCracken *et al* 2002, George and McCracken 2003). However, to date there is no real understanding reported in the literature regarding why nutritive value should vary and no single chemical or physical parameter can be identified as the main contributor of variation (Ball *et al* 2013b). One of the objectives of this current study was to investigate the effect of moisture content at harvest on broiler performance as there has been limited work conducted in this area. It was found that as harvest wheat moisture content increased, broiler LWG increased. There are two possible explanations for this effect. Firstly, the drying process of the wheat prior to storage may have improved the nutritive value of the wheat (Afsharmanesh *et al* 2008). Unfortunately, no information was obtained on the wheat drying conditions so it cannot be conclusively stated that the drying process affected the nutritive value and more work is needed in this area. However, Lawrence *et al* (1986) have shown that increasing starch gelatinisation through cooking can increase digestibility and thus improve performance but this effect was observed on milled wheat and not whole wheat. O'Neill *et al* (2011) suggested that higher moisture content of wheat may be due to an early harvest date which would result in higher levels of cereal amylase and thus starch digestibility in broilers – and not due to the drying process *per* se. However, these workers were unable to conclusively state if the positive effect on starch digestibility was related to higher levels of cereal amylase. The other possible explanation for the increase in LWG may be as a result of an increase in sprouting of the high moisture content wheat which may have produced some nutritional benefits. Chavan and Kadam (1989) stated that sprouting of grain increased the activities of endogenous enzymes thus breaking down protein, carbohydrates and fat to more simple and digestible components. There have been many studies which have shown that sprouting increases the lysine content of the grain through the action of proteolytic enzymes (e.g. Chung *et al* 1989 and Koehler *et al* 2007). Indeed, the theory behind the use of exogenous enzyme supplementation is to mimic the early sprouting process of wheat to breakdown protein and non-starch polysaccharides thus improving the nutritive value. Obviously, there are extremely negative consequences of advanced sprouting on the nutritive value of the grain in the loss of starch (energy), mould growth and the economic cost of drying. In the current study no significant relationship was observed between wheat moisture content at harvest and any chemical parameter. In addition, the wheat samples were not assessed for the level of sprouting and it is difficult to draw firm conclusions without further research.

Another objective of this study was to investigate the effect of microdochium content on performance. This is a novel area and one which does not appear to have been focussed on previously. The assumption that increasing microdochium content would adversely affect bird performance appears to have not been supported as increasing microdochium content resulted in an improvement in feed efficiency. However, the relationship for overall FCR was not strong (R^2=0.19) but it can be concluded that there was no evidence of an adverse effect on microdochium.

Overall, the effect of the mycotoxins DON and ZON, were variable and difficult to explain. This is in keeping with previous research (Andretta *et al* 2011) and while it is accepted mycotoxins reduce performance it is difficult to qualify their effect. A much larger dataset is required to achieve conclusive results. It was an objective of this study to investigate the use of NIR to predict mycotoxin level but as there were variable and confusing effects on performance it was decided that the dataset was not large enough to explore this area effectively. Also, Ochratoxin A was detected in only three of the samples and ochratoxin B detected in only one and it was therefore not possible to investigate their effect on performance.

In the Moy Park trial, wheat sample did not result in any significant difference in performance. This was somewhat unexpected as the wheat samples differed in chemical composition, source of production and some were “new” season wheat. The fact that the diets produced were isonitrogenous and isoenergetic may have masked the effect of wheat sample. However, the diets used in the AFBI trial were also isonitrogenous and isoenergetic and significant differences attributable to wheat sample were observed. It would be interesting to determine protein and essential amino acid digestibilities and the actual AME of both sets of diets to ascertain if the actual values are in keeping with the calculated values. Performance differences due to wheat have been observed in the industry even though diets are formulated to be isonitrogenous and isoenergetic. Furthermore, while there were no significant differences, there was a large range in FCR (1.48 to 1.67), which would result in substantial financial differences. As FCR is a major driver in the cost of producing broiler meat it would useful to have a tool which helps the broiler nutritionist better predict performance on different wheats.

5.3 The use of NIRS to predict microdochium content

There are no other studies reported in the literature which uses NIRS to predict microdochium content. This study found that the strength of relationship between actual and predicted microdochium content was reasonably robust as it is accepted that 1-VR of greater than 0.75 indicates that a strong calibration has been achieved. However, errors associated with the prediction need to be to considered in order to decide the value of the prediction equation. For microdochium prediction, the errors (SECV, SEP and % of means) were reduced when the dataset was split into low, medium and high microdochium content but they were still unacceptably high, as were the RPD values. However, this initial work shows that NIRS may have potential in predicting microdochium content but the dataset needs to be increased to improve the accuracy.

5.4 The use of NIRS to predict broiler performance

Black (2008) reported an unsatisfactory prediction equation for broiler feed intake in a large scale Australian study and establishing accurate prediction equations for broiler performance has proved difficult. For the first time, this study has produced NIRS equations to predict feed intake, LWG and feed efficiency of wheat-based diets validated under commercial conditions. The relationships between actual and predicted performance were strong (R^2>0.8) and importantly, the errors were low and RPD values good indicating accurate and repeatable prediction. These equations will be of great benefit to the broiler industry as a proven, rapid and accurate means of prediction of broiler performance has been developed. However, it must be noted that the relationship between predicted and actual performance of broilers housed under completely commercial conditions was weaker than when the entire dataset was considered. This indicates that more samples should be used to test the equations before they are used commercially.

5.5 Conclusions

1. Specific weight is not a good predictor of wheat chemical composition.
2. There was no relationship between specific weight and broiler performance.
3. Formulating diets from different wheat samples resulted in variable performance across all trials. This highlights the need for an accurate predictor of wheat quality.
4. Supplementary enzyme improved the feeding quality of diets based on wheat samples with lower specific weight and soft endosperms. (Harper Adams University – see Appendix 3).
5. Wheat with a higher moisture content at harvest resulted on better bird performance – perhaps due to the beneficial effects of limited sprouting or the subsequent drying process. More research is required in this area.
6. NIRS has the potential to predict wheat microdochium content but the errors associated with the prediction are high. A larger dataset is needed to develop the equations.
7. Strong prediction equations were developed using data from birds kept under commercial conditions. These equations can accurately predict how birds will perform when offered a diet-based on a particular wheat.
8. Using the prediction equations to compare actual and predicted performance under commercial conditions within Moy Park resulted in a somewhat weaker but still acceptable relationship. The prediction equations should be further strengthened using additional samples and could then be made commercially available.

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8. Appendices

8.1 Appendix 1

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**
Wheat	67.37	67.59	67.66	67.68	67.68	67.71	**67.69**	69.77	68.92	70.00	69.79	69.46	69.64	68.22	67.00	65.00	69.00	69.71	68.79	67.71
Soybean meal	20.42	20.00	20.00	20.00	20.00	20.00	20.00	18.00	18.91	17.69	18.02	18.16	18.22	19.50	20.76	23.50	19.00	18.18	19.00	20.00
Full fat soya	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Soya oil	4.30	4.32	4.31	4.31	4.31	4.31	4.31	4.14	4.17	4.17	4.13	4.23	4.11	4.27	4.30	3.88	4.24	4.11	4.22	4.31
Salt	0.14	0.12	0.14	0.13	0.13	0.14	0.14	0.12	0.12	0.11	0.12	0.11	0.12	0.13	0.23	0.15	0.13	0.12	0.13	0.14
Sodium Bicarbonate	0.34	0.45	0.44	0.44	0.44	0.43	0.44	0.46	0.37	0.47	0.45	0.47	0.37	0.44	0.21	0.35	0.44	0.37	0.44	0.44
DL Methionine	0.23	0.24	0.24	0.23	0.23	0.22	0.22	0.23	0.23	0.23	0.23	0.24	0.24	0.23	0.25	0.00	0.00	0.23	0.21	0.22
Lysine HCl	0.02	0.07	0.02	0.03	0.03	0.01	0.02	0.08	0.08	0.10	0.06	0.11	0.08	0.04	0.02	0.00	0.03	0.09	0.04	0.02
Threonine	0.04	0.07	0.05	0.04	0.04	0.04	0.04	0.05	0.06	0.08	0.05	0.07	0.07	0.03	0.10	0.00	0.02	0.04	0.03	0.02
L-Tryptophan	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Limestone	1.01	1.01	1.01	1.01	1.01	1.01	1.01	1.02	1.01	1.02	1.02	1.02	1.02	1.01	1.00	1.00	1.01	1.02	1.01	1.01
Mono Dical Phos	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.61	0.60	0.61	0.61	0.61	0.61
Mins and vits	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Phytase	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02

8.2 Appendix 2

	Starter Crumb (Maxiban)						Grower Pellet (Maxiban)						Finisher Pellet (Elancoban)						Withdrawal Pellet					
	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6
Danish Old Crop Wheat	58.697	0	0	0	0	0	60.072	0	0	0	0	0	66.778	0	0	0	0	0	66.048	0	0	0	0	0
Romanian New Crop Wheat	0	61.592	0	0	0	0	0	62.693	0	0	0	0	0	70.632	0	0	0	0	0	69.607	0	0	0	0
UK Old Crop Wheat	0	0	59.582	0	0	0	0	0	61.057	0	0	0	0	0	67.943	0	0	0	0	0	67.013	0	0	0
Uk New Crop Wheat	0	0	0	58.697	0	0	0	0	0	60.072	0	0	0	0	0	66.982	0	0	0	0	0	66.048	0	0
Irish Wheat	0	0	0	0	60.567	0	0	0	0	0	61.932	0	0	0	0	0	69.047	0	0	0	0	0	68.117	0
Danish New Crop	0	0	0	0	0	57.832	0	0	0	0	0	59.197	0	0	0	0	0	66.227	0	0	0	0	0	65.072
GM Hipro Soya	29.2	26.1	28.3	29.2	27.5	30	21.8	19.3	20.9	21.8	20.1	22.6	18.7	15.2	17.7	18.4	16.7	18.9	16.1	12.8	15.2	16.1	14.2	17
DDGS	2.5	2.5	2.5	2.5	2.5	2.5	7.5	7.5	7.5	7.5	7.5	7.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5
Whole Rapeseed	3	3	3	3	3	3	4.5	4.5	4.5	4.5	4.5	4.5	0	0	0	0	0	0	5.5	5.5	5.5	5.5	5.5	5.5
Oil Blend	2.4	2.6	2.4	2.4	2.2	2.5	2.5	2.3	2.4	2.5	2.3	2.6	4.9	4.4	4.7	4.8	4.6	4.8	3	2.7	2.9	3	2.8	3.1
Limestone	0.85	1	0.85	0.85	0.85	0.85	1	1.05	1	1	1	1	1.05	1.1	1.05	1.05	1.05	1.05	0.9	0.9	0.9	0.9	0.9	0.9
Mono-Dicalcium Phosphate	0.85	0.85	0.85	0.85	0.85	0.85	0.6	0.6	0.6	0.6	0.6	0.6	0.5	0.5	0.5	0.5	0.5	0.5	0.4	0.4	0.4	0.4	0.4	0.4
Vitamin & Mineral Premix	0.7	0.7	0.7	0.7	0.7	0.7	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.5	0.5	0.5	0.5	0.5	0.5
L-Lysine-HCl	0.42	0.5	0.44	0.42	0.46	0.395	0.44	0.5	0.46	0.44	0.485	0.42	0.445	0.525	0.475	0.53	0.495	0.53	0.47	0.545	0.5	0.47	0.52	0.45
DL Methionine	0.395	0.38	0.39	0.395	0.39	0.39	0.305	0.285	0.3	0.305	0.305	0.305	0.29	0.275	0.29	0.375	0.285	0.63	0.285	0.27	0.285	0.285	0.28	0.285
Additive	0.3	0.3	0.3	0.3	0.3	0.3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Soyabean oil	0.3	0	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0	0	0	0	0	0	0	0	0	0	0	0
Sodium Carbonate	0.25	0.35	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.15	0.2	0.15	0.2	0.15	0.2	0.15	0.15	0.15	0.15	0.15	0.15
Salt	0.06	0.03	0.05	0.06	0.04	0.06	0.05	0.028	0.04	0.05	0.03	0.05	0.028	0	0.028	0	0	0	0.028	0	0.028	0.028	0	0.03
Threonine	0.045	0.065	0.055	0.045	0.06	0.04	0.05	0.06	0.06	0.05	0.065	0.045	0.025	0.035	0.03	0.03	0.04	0.03	0.085	0.095	0.09	0.085	0.1	0.08
NSP Enzyme	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023	0.023
Phytase Enzyme	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Protein	21.75	21.75	21.75	21.75	21.75	21.75	20	20	20	20	20	20	18	18	18	18	18	18	17.75	17.75	17.75	17.75	17.75	17.75
Energy Kcal/lb	1385	1385	1385	1385	1385	1385	1425	1425	1425	1425	1425	1425	1455	1455	1455	1455	1455	1455	1460	1460	1460	1460	1460	1460
Calcium	0.9	0.9	0.9	0.9	0.9	0.9	0.85	0.856	0.85	0.85	0.85	0.85	0.8	0.81	0.8	0.8	0.8	0.8	0.75	0.75	0.75	0.75	0.75	0.75
Av Phoshorus	0.453	0.452	0.453	0.45	0.45	0.452	0.422	0.422	0.422	0.425	0.425	0.425	0.391	0.392	0.392	0.391	0.392	0.391	0.38	0.38	0.38	0.38	0.38	0.38
Salt	0.293	0.29	0.29	0.293	0.287	0.284	0.303	0.302	0.3	0.303	0.299	0.296	0.281	0.28	0.291	0.28	0.28	0.28	0.293	0.29	0.303	0.293	0.281	0.287

	Starter Crumb (Maxiban)						Grower Pellet (Maxiban)						Finisher Pellet (Elancoban)						Withdrawal Pellet					
	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6
Sodium	0.18	0.21	0.18	0.18	0.172	0.18	0.185	0.176	0.181	0.185	0.18	0.185	0.13	0.14	0.13	0.14	0.13	0.14	0.131	0.12	0.131	0.131	0.13	0.132
Total Lysine	1.43	1.43	1.43	1.43	1.43	1.43	1.3	1.3	1.3	1.3	1.3	1.3	1.17	1.16	1.17	1.22	1.17	1.23	1.17	1.17	1.18	1.17	1.17	1.17
Total Methionine+Cysteine	1.07	1.07	1.07	1.07	1.07	1.07	0.97	0.97	0.97	0.97	0.97	0.97	0.89	0.89	0.89	0.97	0.88	1.21	0.89	0.89	0.89	0.89	0.89	0.89
Total Threonine	0.92	0.92	0.92	0.92	0.92	0.92	0.84	0.84	0.84	0.84	0.84	0.84	0.78	0.77	0.77	0.78	0.78	0.78	0.78	0.77	0.78	0.78	0.78	0.78

8.3 Appendix 3

Growth performance of broilers fed diets based on six different wheat cultivars with and without enzyme (Econase XT25 P) supplementation

A Study Report for:

AHDB Cereals & Oilseeds project RD-2012-3805

“Development of an accurate means to predict the nutritive value of wheat for broilers and an investigation of the effect of harvest moisture content of wheat on broiler performance”

Prepared by:

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1. QUALITY STATEMENT

Harper Adams University has a documented quality policy and is committed to providing the highest possible quality of service to its clients in the provision of education, research, and advisory and consultancy services.

Procedures, documentation, equipment and records were examined in order to assure that the study was conducted in accordance with the regulations specified herein and with the protocol and relevant methods, in collaboration with the trial monitor.

The study was subjected to scrutiny and approval by Harper Adams University Animal Experiments Committee.

Dr Vasil Pirgozliev – Research Site Study Investigator

Signed: ____

Dated: ____

2. STUDY RESPONSIBILITY

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3. SUMMARY

Twelve wheat-based diets, based on six different wheat cultivar samples with and without enzyme (Econase XT 25 P) supplementation, were fed to Ross 308 male broilers from 0 to 42 days of age.

One thousand nine hundred and twenty chicks were allocated to 96 floor pens, giving twenty birds in each pen. Each diet was replicated 8 times in a randomised complete block design. There were three dietary phases and diets were changed at 11 and 27d age. Diets were fed as mash from 0 to 11 d age, and as pellets from 11d to 27d, and from 27d to 42 d age. Feed and water were offered *ad libitum* through entire experimental period.

Broiler growth performance was compared statistically by ANOVA using a 6 x 2 factorial arrangement of treatments. The main effects are the wheat cultivar and the enzyme (with and without) used. In all instances, differences are reported as significant at $P < 0.05$. Tendencies towards significance ($0.05 < P < 0.1$) are also included in the report.

The results form laboratory analyses were in the expected range. Supplementary Econase XT25 P improved the feeding quality of diets based on wheat samples with lower specific weight and soft endosperm compared to wheat samples with relatively hard endosperm and high specific weight. The results suggest that supplementary Econase may be more effective in diets containing poor quality wheat.

4. STUDY OBJECTIVE

The objectives of the study were to evaluate the effect of six wheat cultivar samples, with and without enzyme, when fed to broiler chickens from 0 to 42 days of age on bird feed intake, growth and feed efficiency.

5. MATERIALS

Table 1. Study Animals and Husbandry Conditions

Number of treatments	12
Replicate pens per treatment	8
Birds per replicate pen	20
Species of bird	Broiler chicken
Breed of bird	Ross 308 hybrid
Sex of birds	Males
Range of trial	0 - 42 days
Diet form (pellet/meal)	Meal
Coccidiostat	Mareks disease and IB in the hatchery
Age at which birds are weighed	0, 11, 27 and 42 days
Age at which feed is weighed	0, 11, 27 and 42 days
Variables measured	Bird weight, feed consumption and FCR
Inspections	The health and welfare of all birds were inspected daily. No problems were observed.
In the event of mortality, health, welfare or any unforeseen issues or concerns - especially if there is a high early mortality (e.g. chick mortality (including culls) reaches 3% within 72 hours of placement)	The mortality was less than 1% (17 birds in total) and did not relate to dietary treatments.
Stocking density (birds/m^2)	Less than 32kg/m^2
Lighting programme	16 hours light per day
House temperature programme	31 - 20°C
House humidity programme	N/A
Vaccination programme	Commercial hatchery programme
Ventilation – Air changes/hr	N/A

Table 2. Test products

Product	Name	Source
NSP enzyme	Econase XT	ABVista

In the enzyme based treatments an equivalent of 100 g enzyme mixture / tonne diet were mixed (100g/tonne 16,000 enzyme activity units/kg).

Table 3. Description of test wheat

Wheat name	DM (g/kg)	GE (MJ/kg DM)	CP (g/kg DM)	Sp. Weight (kg/hl)	Endosperm Hardness	In vitro viscosity mPa.s
SA358	914	18.03	128.4	81.8	62.60	8.28
SA359	919	17.87	105.9	74.0	41.56	8.81
SA360	924	17.93	116.7	71.2	31.62	10.15
Solstice	922	17.95	126.9	70.9	36.18	9.23
Brockton	925	17.84	110.6	66.0	33.14	10.5
Neil Furniss	920	17.80	109.6	57.0	21.97	11.35

The experimental wheat samples were provided by AHDB project RD-2012-3805 “Development of an accurate means to predict the nutritive value of wheat for broilers and an investigation of the effect of harvest moisture content of wheat on broiler performance”. The enzyme (Ecoanse XT25 P) was provided by AB Vista.

Table 4. Diet related information

Diet related information	
Date of manufacture	May 2014
Treatment Batch size / kg	Various: 60 kg mash of each starter diet; 270 kg pellets of each grower diet; 650 kg pellets of each finisher diet.
Description of mixer (Size, model, mixing time to be include in the report)	Target feeds Ltd
Diet form	**Mash & Pellet**
Samples collected per diet/quantity per sample	A sample from each diet (approximately 200g) was send for enzyme analysis to: Mr Noel Sheehan; ESC Ltd Unit 6 Innovation and Technology Centre Tredomen Park Ystrad Mynach Hengoed CF82 7FQ Wales
Diet / Water availability	*Ad libitum / Ad libitum*
Diet phases	One phase
Analysis of diet requested	Moisture, Crude protein, Minerals
Coccidiostat used	In Starter and Grower diets only
Growth promoter used	No
Mycotoxin binder used	No

All diets were manufactured in Target Feeds, under the direct supervision of Mr. Ian Hollows. Feeds were stored in a cool dry place before the experiment. Diets were prepared without the inclusion of any growth promoter, antibiotic or coccidiostat and were analysed for gross energy, dry matter, protein, and minerals by Harper Adams University. Parallel samples were analysed for various parameters and enzyme activities by AB Vista (samples sent to Mr Noel Sheehan).

Product Storage Conditions: The products were stored in a cool (<20°C), dry place, protected from moisture until mixed with the trial diets.

Table 5. Experimental diets formulation

Ingredient	Composition g/kg		
	Starter 0-10d	Grower 11-26d	Finisher 27-42d
Test wheat	670.0	670.0	670.0
Soya bean meal (CP=48%)	226.1	172.22	140.0
Full fat Soya meal	48.9	98.85	139.0
Soya oil	10	20.0	20.0
Lysine	3.0	3.0	1.4
Methionine	4.4	4.1	3.1
Monocalcium phosphate	15.5	11.7	8.5
Limestone	15.0	13.5	11.0
Salt	3.3	3.0	3.0
Vitamin/mineral premix	4.0	4.0	4.0
Calculated composition			
ME (MJ/kg)	12.43	12.95	13.18
Protein (g/kg)	212.7	204.0	200.4
Lysine (g/kg)	1.35	13.0	11.3
Met + Cys (g/kg)	9.8	9.4	8.6
Calcium (g/kg)	9.9	8.5	7.0
Phosphorus av (g/kg)	4.8	4.0	3.3
Sodium (g/kg)	1.9	1.7	1.7

Starter diets were fed as mash and the grower and finisher diets were fed as pellets.

Table 6. Treatment identification

Treatment ID	Wheat	Enzyme
Diet 1	SA358	No
Diet 2	SA359	No
Diet 3	SA360	No
Diet 4	Solstice	No
Diet 5	Brockton	No
Diet 6	Neil Furniss	No
Diet 7	SA358	Ecoanse XT25 P (100g/tonne 16,000u/kg)
Diet 8	SA359	Ecoanse XT25 P (100g/tonne 16,000u/kg)
Diet 9	SA360	Ecoanse XT25 P (100g/tonne 16,000u/kg)
Diet 10	Solstice	Ecoanse XT25 P (100g/tonne 16,000u/kg)
Diet 11	Brockton	Ecoanse XT25 P (100g/tonne 16,000u/kg)
Diet 12	Neil Furniss	Ecoanse XT25 P (100g/tonne 16,000u/kg)

Table 7. Numbers of birds, replicates and treatments in the Experiment

No. of treatments	12	Broilers per replicate	20
Replicates per treatment	8	Broilers per treatment	160
Total No. of replicates	96	Total No. of broilers	1920

6. METHODS

6.1. Animal husbandry and dietary analysis

Approximately one thousand nine hundred and sixty (1,960) day-old male Ross 308 chicks were obtained from a commercial hatchery. On arrival one thousand nine hundred and twenty chicks were allocated to 96 floor pens, twenty birds in each pen. Each of the pens has a concrete floor with an area of 1.40 X 1.50 m that was covered with a bedding material (wood shavings). Birds received one of the 12 experimental diets. Feed and water were offered *ad libitum* to birds throughout the experiment.

Each diet was offered to birds in 8 pens in a randomised complete block design. Information on growth and feed intake was obtained from 0 to 42 days of age. The room temperature was

approximately 31ºC, at day-old, and was gradually reduced to 20ºC when birds were 20 days old. A standard lighting programme for broilers was used.
Birds and feed were weighed at the beginning (0 day old) and at the end (11 day old) of the starter phase, at the beginning (11 day old) and at the end (27 day old) of the grower phase and at the beginning (27 day old) and at the end (42 day old) of the finisher phase of the study, and the weight gain and feed efficiency were determined.

Wheat and diets were analysed for energy and nutrients following standard procedures.
Pellet durability was tested by Holmen test.

6.2. Statistical analysis

Statistical analyses were performed using the Genstat statistical software package (Genstat 15th release 3.22 for Windows; Rothamsted, Hertfordshire, UK). Broiler growth performance was compared statistically by analysis of variance using a 6 x 2 factorial arrangement of treatments. The main effects are the wheat cultivar and the enzyme (with and without) used. In all instances, differences are reported as significant at $P < 0.05$. Tendencies towards significance ($0.05 < P < 0.1$) are also included in the report.

7. RESULTS

The results on the dry matter (DM), gross energy (GE), crude protein (CP), Ca and P of the experimental diets are presented on Table 8a, b and c, and were in the expected range. The dietary pellets did not differ in their quality and were with medium durability. Room temperature during study is presented on Appendix 1.

Birds fed diets based on wheat cultivar Solstice tended ($P=0.074$) to consume more feed compared to those fed wheat SA359 (Table 9) for the entire study period. Enzyme supplementation did not influence ($P>0.05$) overall dietary feed intake.
Birds fed diets based on wheat cultivar Solstice tended ($P=0.095$) to have overall greater weight gain compared to those fed wheat SA359 (Table 10). Although Econase supplementation improved ($P<0.001$) dietary feed intake for the first feeding phase, enzyme supplementation did not influence ($P>0.05$) overall dietary feed intake.

The overall final weight of the birds was 2.910 kg (data not included in tables) and in agreement with breeders performance objectives (Aviagen Ltd.).

There was wheat cultivar by Econase interaction (P<0.001) for the overall feed conversion ratio (FCR) as half of the wheats (Brockton, NF and SA360), benefitted from enzyme supplementation, although the rest (SA358, SA359 and Solstice) did not (Table 11).

The results for mortality corrected FCR was similar to those for FCR (Table 12). Some of the diets, e.g. based on wheat obtained from Brockton (wheat with relatively soft endosperm and high *in vitro* viscosity; Table 3), benefitted from Econase supplementation as the MFCR was reduced (P<0.001), although diets based on SA358, SA359 tended to increase MFCR as a result of enzyme supplementation. The FCR and MFCR values are based on dry matter feed intake, so to obtain the ration on as fed basis, the values need to be divided by the dry matter of the diet.
The results suggest that wheat cultivar samples with lower specific weight and soft endosperm benefitted from Econase supplementation compared to wheat samples with relatively hard endosperm and high specific weight.

Table 8a. Dry matter (DM) and pellet quality (Holmen test, HT) of the experimental diets

Treatment	Wheat	Enzyme	DMs (g/kg)	DMg (g/kg)	DMf (g/kg)	HTg (%)	HTf (%)
1	SA358	no	880	881	884	88.2	87.8
2	SA359	no	878	876	885	84.6	85.6
3	SA360	no	876	876	883	83.0	87.6
4	Solstice	no	878	878	883	83.2	86.2
5	Brockton	no	881	875	880	86.2	79.2
6	Neil Furniss	no	880	873	880	84.8	84.8
7	SA358	yes	885	878	880	82.4	82.8
8	SA359	yes	882	876	879	82.6	84.2
9	SA360	yes	880	872	878	83.6	84.8
10	Solstice	yes	884	874	883	85.2	85.4
11	Brockton	yes	883	877	880	84.2	86.0
12	Neil Furniss	yes	880	872	879	86.0	82.6

Table 8b. Crude protein (CP) and gross energy (GE) of the experimental diets

Treatment	Wheat	Enzyme	CPs (g/kg)	CPg (g/kg)	CPf (g/kg)	GEs (MJ/kg)	GEg (MJ/kg)	GEf (MJ/kg)
1	SA358	no	325.0	325.9	310.9	16.08	16.76	16.88
2	SA359	no	325.1	283.6	306.6	16.26	16.62	17.03
3	SA360	no	322.8	291.0	301.6	16.35	16.68	16.92
4	Solstice	no	372.6	312.7	319.2	16.47	16.70	17.09
5	Brockton	no	346.8	284.6	297.9	15.98	16.68	16.89
6	Neil Furniss	no	365.6	313.8	297.5	16.03	16.54	16.90
7	SA358	yes	310.8	309.5	322.2	15.93	16.65	16.92
8	SA359	yes	311.9	302.3	295.6	15.93	16.58	16.90
9	SA360	yes	281.4	305.5	287.2	15.69	16.66	16.85
10	Solstice	yes	322.6	327.1	331.8	15.88	16.63	16.95
11	Brockton	yes	354.7	304.7	297.0	16.05	16.62	16.92

12	Neil Furniss	yes	296.4	291.1	310.8	15.89	16.59	16.98

Table 8c. Dietary P and Ca contents (g/kg DM)

Treatment	Wheat	Enzyme	P s	P g	P f	Ca s	Ca g	Ca f
1	SA358	no	10.1	8.16	8.00	15.3	12.45	11.70
2	SA359	no	9.8	8.09	7.64	14.3	11.95	11.25
3	SA360	no	10.0	8.86	9.30	14.9	12.00	12.45
4	Solstice	no	9.5	8.49	7.25	13.2	12.15	8.99
5	Brockton	no	9.1	8.82	7.72	12.3	12.15	10.70
6	Neil Furniss	no	11.1	8.39	8.50	18.4	12.00	11.60
7	SA358	yes	9.9	8.06	6.92	15.7	11.90	8.53
8	SA359	yes	10.2	8.82	7.22	15.3	12.55	9.48
9	SA360	yes	10.1	8.39	8.51	14.4	11.55	12.00
10	Solstice	yes	9.9	8.07	8.31	12.9	11.70	12.60
11	Brockton	yes	10.1	8.33	8.31	14.8	11.80	12.75
12	Neil Furniss	yes	10.3	9.11	8.25	15.4	13.25	11.70

s – starter diets; g – grower diets; f – finisher diets

Table 9. Effect of the experimental wheat-based diets with and without Econase XT25 P supplementation on daily feed intake (FI) expressed on dry matter (DM) basis during different growing phases

Treatment factor	FI 0-11d (g DM/b/d)	FI 11-27d (g DM/b/d)	FI 27-42d (g DM/b/d)	FI 0-42d (g DM/b/d)
Wheat #				
Brockton	20.58	79.40	168.00	96.72
NF	19.86	76.80	169.61	96.39
SA358	20.56	78.29	161.12	93.73
SA359	20.34	75.82	162.31	93.50
SA360	20.78	77.64	163.85	94.69
Solstice	20.89	80.85	167.51	97.37
SEM	0.424	1.360	1.876	1.133
Econase				
No	20.20	77.35	166.50	95.36
Yes	20.80	78.92	164.30	95.43
SEM	0.245	0.785	1.083	0.654
Brockton	19.86	80.43	170.59	98.03
NF	18.92	74.95	170.96	95.90
SA358	20.74	78.66	161.42	93.88
SA359	20.40	73.14	161.95	92.30
SA360	20.00	78.05	166.77	95.63
Solstice	21.30	78.85	167.29	96.42
Brockton + Econase	21.29	78.38	165.41	95.40
NF + Econase	20.80	78.64	168.25	96.87
SA358 + Econase	20.38	77.91	160.82	93.58
SA359 + Econase	20.29	78.51	162.67	94.69
SA360 + Econase	21.55	77.24	160.94	93.75
Solstice + Econase	20.49	82.85	167.73	98.31
SEM	0.600	1.923	2.653	1.602

Probabilities of statistical differences (P)				
Wheat	0.589	0.129	0.008	0.074
Econase	0.089	0.163	0.156	0.939
Diets x Econase	0.112	0.263	0.720	0.545
CV%	8.3	7.0	4.5	4.8

P - significance between treatments determined by ANOVA. SEM - standard errors of means; CV% - the coefficient of variation.

Table 10. Effect of the experimental wheat-based diets with and without Econase XT25 P supplementation on daily weight gain (WG) during different growing phases

Treatment factor	WG 0-11d (g/b/d)	WG 11-27d (g/b/d)	WG 27-42d (g/b/d)	WG 0-42d (g/b/d)
Wheat #				
Brockton	14.83^{c}	67.14^{bc}	106.17^{b}	68.6
NF	13.4^{a}	63.40^{a}	110.13^{c}	68.20
SA358	14.76^{c}	67.7^{c}	101.91^{a}	67.35
SA359	13.60^{ab}	64.53^{ab}	105.49^{b}	67.03
SA360	14.55^{bc}	66.39^{bc}	106.39^{b}	68.33
Solstice	14.97^{c}	68.70^{c}	108.55^{bc}	70.14
SEM	0.349	0.979	1.266	0.784
Econase				
No	13.65	66.59	105.70	67.95
Yes	15.07	66.06	107.18	68.61
SEM	0.202	0.565	0.731	0.453
Brockton	14.32	68.57	103.74	68.22
NF	12.55	62.4	109.64	67.40
SA358	14.43	69.4	102.22	68.05
SA359	12.51	63.26	105.94	66.42
SA360	13.91	67.32	105.73	68.32
Solstice	14.16	68.57	106.90	69.31
Brockton + Econase	15.33	65.71	108.59	69.03
NF + Econase	14.37	64.39	110.63	69.00
SA358 + Econase	15.10	66.15	101.60	66.66
SA359 + Econase	14.68	65.81	105.04	67.65
SA360 + Econase	15.18	65.47	107.04	68.34
Solstice + Econase	15.78	68.84	110.19	70.97
SEM	0.494	1.384	1.791	1.109
Probabilities of statistical differences (P)				
Wheat	0.006	0.002	<0.001	0.095

Econase	<0.001	0.509	0.155	0.309
Diets x Econase	0.689	0.161	0.569	0.733
CV%	9.7	5.9	4.8	4.6

P - significance between treatments determined by ANOVA. SEM - standard errors of means; CV% - the coefficient of variation. Superscripts ^{a,b,c} within columns indicate significance – those without a common superscript are significantly different (<0.05).

Table 11. Effect of the experimental wheat-based diets with and without Econase XT25 P supplementation on feed conversion ratio (FCR) expressed on dry matter intake during different growing phases

Treatment factor	FCR 0-11d	FCR 11-27d	FCR 27-4211d	FCR 0-42d
Wheat #				
Brockton	1.392	1.183^{b}	1.586	1.410
NF	1.496	1.210^{c}	1.540	1.413
SA358	1.398	1.155^{a}	1.581	1.392
SA359	1.519	1.174^{b}	1.539	1.395
SA360	1.432	1.170^{ab}	1.541	1.386
Solstice	1.405	1.176^{b}	1.544	1.388
SEM	0.0358	0.0056	0.0106	0.0065
Econase				
No	1.495	1.161	1.577	1.404
Yes	1.385	1.194	1.534	1.391
SEM	0.0207	0.0032	0.0061	0.0038
Brockton	1.390	1.173	1.648	1.438
NF	1.537	1.199	1.559	1.423
SA358	1.443	1.133	1.580	1.380
SA359	1.654	1.155	1.530	1.390
SA360	1.441	1.160	1.578	1.400
Solstice	1.507	1.150	1.565	1.391
Brockton + Econase	1.395	1.192	1.524	1.382
NF + Econase	1.456	1.221	1.521	1.404
SA358 + Econase	1.353	1.177	1.583	1.404
SA359 + Econase	1.385	1.193	1.549	1.400

SA360 + Econase	1.422	1.180	1.503	1.372
Solstice + Econase	1.303	1.202	1.523	1.385
SEM	0.0507	0.0079	0.0151	0.0092
Probabilities of statistical differences (P)				
Wheat	0.053	<0.001	0.002	0.012
Econase	<0.001	<0.001	<0.001	0.022
Diets x Econase	0.062	0.166	<0.001	<0.001
CV%	10	1.9	2.7	1.9

P - significance between treatments determined by ANOVA. SEM - standard errors of means; CV% - the coefficient of variation. Superscripts ^{a,b,c} within columns indicate significance – those without a common superscript are significantly different (<0.05).

Table 12. Effect of the experimental wheat-based diets with and without Econase XT25 P supplementation on mortality corrected feed conversion ratio (MFCR) expressed on dry matter intake during different growing phases

Treatment factor	FCR 0-11d	FCR 11-27d	FCR 27-4211d	FCR 0-42d
Wheat #				
Brockton	1.392	1.184^{b}	1.586	1.415
NF	1.496	1.210^{c}	1.540	1.413
SA358	1.400	1.161^{a}	1.583	1.400
SA359	1.519	1.174^{ab}	1.541	1.396
SA360	1.432	1.172^{ab}	1.541	1.390
Solstice	1.405	1.179^{b}	1.544	1.391
SEM	0.0358	0.0055	0.0106	0.0058
Econase				
No	1.496	1.165	1.577	1.408
Yes	1.385	1.195	1.535	1.394
SEM	0.0207	0.0032	0.0061	0.0034
Brockton	1.390	1.175	1.648	1.441
NF	1.537	1.199	1.559	1.423
SA358	1.448	1.143	1.580	1.392
SA359	1.654	1.155	1.530	1.390

SA360	1.441	1.163	1.578	1.405
Solstice	1.507	1.156	1.565	1.397
Brockton + Econase	1.395	1.193	1.525	1.389
NF + Econase	1.456	1.221	1.521	1.404
SA358 + Econase	1.353	1.179	1.586	1.409
SA359 + Econase	1.385	1.193	1.552	1.401
SA360 + Econase	1.422	1.182	1.503	1.374
Solstice + Econase	1.303	1.202	1.523	1.385
SEM	0.0506	0.0077	0.0150	0.0082
Probabilities of statistical differences (P)				
Wheat	0.055	<0.001	0.001	0.006
Econase	<0.001	<0.001	<0.001	0.004
Diets x Econase	0.062	0.326	<0.001	<0.001
CV%	9.9	1.9	2.7	1.7

P - significance between treatments determined by ANOVA. SEM - standard errors of means; CV% - the coefficient of variation. Superscripts ^{a,b,c} within columns indicate significance – those without a common superscript are significantly different (<0.05).

8. CONCLUSIONS

Supplementary Econase XT25 P improved the feeding quality of diets based on wheat samples with lower specific weight and soft endosperm compared to wheat samples with relatively hard endosperm and high specific weight.

Appendix 1. Room minimum and maximum temperatures during the study

Age of birds, days	Max. T˚C	Min. T˚C	Age of birds, days	Max. T˚C	Min. T˚C
0	33.1	32.9			
1	32.6	32.1	**22**	25.4	23.8
2	32.6	32.1	**23**	25.5	22.8
3	32.7	32.2	**24**	24.6	22.5
4	32.6	31.3	**25**	23.4	22
5	32.1	31.1	**26**	23.4	21.5
6	31.5	30.7	**27**	22.7	20.8
7	31.2	29.8	**28**	22.3	20.5
8	30.9	29.4	**29**	21.9	20.1
9	30.2	28.8	**30**	21.9	19.4
10	29.3	28.3	**31**	23.4	19.7
11	28.9	27.6	**32**	23.9	19.4
12	28.9	27.2	**33**	22.7	19.7
13	27.9	26.7	**34**	22.7	19.7
14	27.6	26.6	**35**	25.5	19.7
15	27.6	26.2	**36**	24.2	19.3
16	26.8	25.8	**37**	24.3	19
17	26.5	25.1	**38**	23.6	19.1
18	25.7	24.7	**39**	23.5	19.2
19	25.7	24.7	**40**	24.2	19.1
20	25.7	24.7	**41**	24.2	19.3
21	24.8	23.7	**42**	25.5	19.1